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# The Synthesis of α-Amino Acids Using the Regiospecific Ring Opening of α-Amino-β-Lactones and the Preparation of some Thalidomide Analogs

by

John Channon Gerald Drover

(C)

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

Department of Chemistry

EDMONTON, ALBERTA

Fall 1986

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.....

Supervisor

.....

.....

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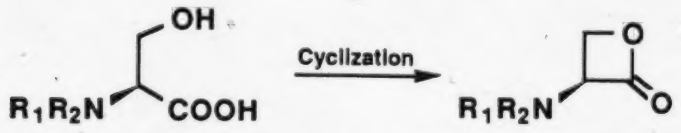
Date September 30, 1986

To Jim and Annie

## Abstract

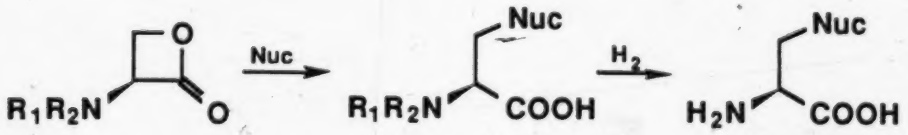
A new method for the synthesis of α-amino acids has been developed. This could potentially lead to substituted derivatives of glutamic acid, and hence to the syntheses of analogs of the teratogen thalidomide (**36**). N-Protected serines **58** and **72** are cyclized without racemization to N-protected α-amino β-lactones **55** and **64** using modified Mitsunobu conditions ($Ph_3P$, dimethyl azodicarboxylate, -78 °C).




**58** $R_1 = H, R_2 = OCOCH_2Ph$ — **55** $R_1 = H, R_2 = OCOCH_2Ph$

**72** $R_1 = CH_2Ph, R_2 = OCOCH_2Ph$ — **64** $R_1 = CH_2Ph, R_2 = OCOCH_2Ph$

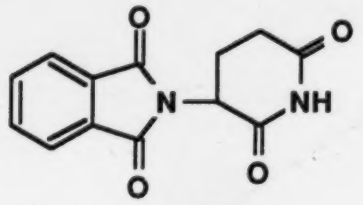
Treatment of β-lactone **64** with a variety of halogen, oxygen, sulfur, nitrogen, or organocuprate based nucleophiles gives good yields (51-99%) of N-protected β-substituted alanines.




**97** Nuc = $CH_3$
**98** = Ph
**99** = $(CH_3)_3$

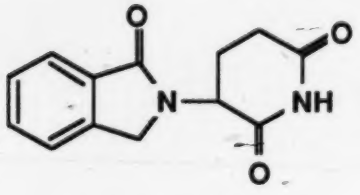
Hydrogenation of these N-protected β-substituted alanines affords the optically active free α-amino acids **97-99** in high yield (79-99%).

Thalidomide (**36**) is a potent human teratogen. It was prescribed as a safe sedative-hypnotic and in early 1960 became a commonly prescribed antiemitic for pregnant women. A year later it was linked to the observation of increased



**36**

numbers of congenital birth defects. A number of theories have been proposed to explain the mechanism of action of this drug but these biochemical details remain elusive. Thalidomide analogs **37**, **42**, **44**, and **45** were prepared to further probe the structure activity relationships of this drug.



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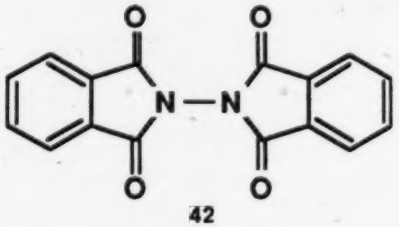
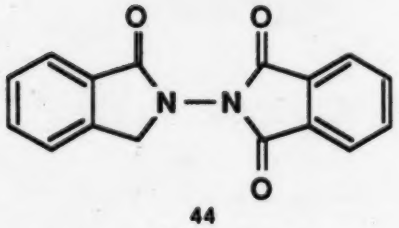
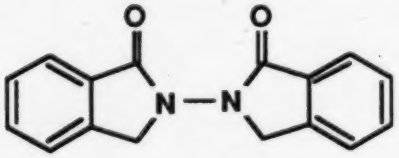


**42**



**44**



**45**

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## Table of Contents


Page

I. Introduction ..... 1

A.) Some Recent Methods for Amino Acid Synthesis ..... 1

B.) Nitrogen-Substituted β-Lactones as Possible Precursors for Amino Acids ..... 9

C.) Thalidomide and its Background ..... 17

II. Results and Discussion ..... 23

III. Experimental ..... 50

IV. References ..... 88

## List of Tables


| Table | | Page |
|---|---|---|
| 1 | Reaction of N-Benzyl-N-Benzyloxycarbonyl-α-Amino Serine β-Lactone **64** with Non-Carbon Nucleophiles | 33 |
| 2 | Reaction of N-Benzyl-N-Benzyloxycarbonyl α-Amino Serine β-Lactone **64** with Organocuprates | 35 |
| 3 | Gas Chromatographic Retention Times of 2' Isomers of Camphanamide Methyl Esters of α-Aminobutyric Acid | 41 |
| 4 | Gas Chromatographic Retention Times of 2' Isomers of Camphanamide Methyl Esters of Phenylalanine | 41 |

## List of Abbreviations

Ac..... Acetyl

Bn..... Benzyl (phenylmethyl)

Bu..... Butyl

DCC..... Dicyclohexylcarbodiimide

DMAP..... 4-Dimethylaminopyridine

DMF..... N,N-Dimethylformamide

EDTA..... Ethylenediaminetetraacetic acid

Et..... Ethyl

IR..... Infrared

LAH..... Lithium aluminum hydride

LDA..... Lithium diisopropyl amide

Me..... Methyl

MPLC..... Medium pressure liquid chromatography

MS..... Mass spectra

NMR..... Nuclear magnetic resonance

*o*..... Ortho

Ph..... Phenyl

Pr..... Propyl

$R_f$..... Retardation factor

rt..... Room temperature

THF..... Tetrahydrofuran

TLC..... Thin layer chromatography

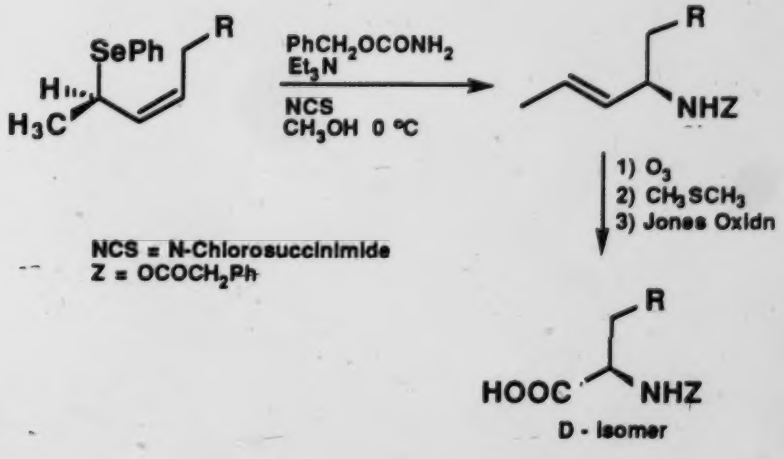
Z..... Benzyloxycarbonyl

# INTRODUCTION

Optically active D- or L-amino acids are constituents of a vast number of biologically active products such as hormones, enzymes, peptides, enzyme inhibitors and pharmaceuticals.[1-12] Thus efficient synthetic routes to them have been the subject of intense effort.[8-28] Strict requirements govern the general applicability of any one method. It must be easily performed to give high chemical and optical yields with predictable stereochemistry. If a chiral auxiliary is used it should be easily recoverable and be readily available in both enantiomeric forms. Most methods fall short in at least one of these criteria. This thesis presents the results of a new method for the asymmetric synthesis of amino acids using the regiospecific ring opening of α-amino-β-lactones prepared from the readily available amino acid, serine. Some of the more recent methods which are reviewed below rely on sigmatropic rearrangements, model enzymes, radical couplings or stereospecific hydrogenations and alkylations.

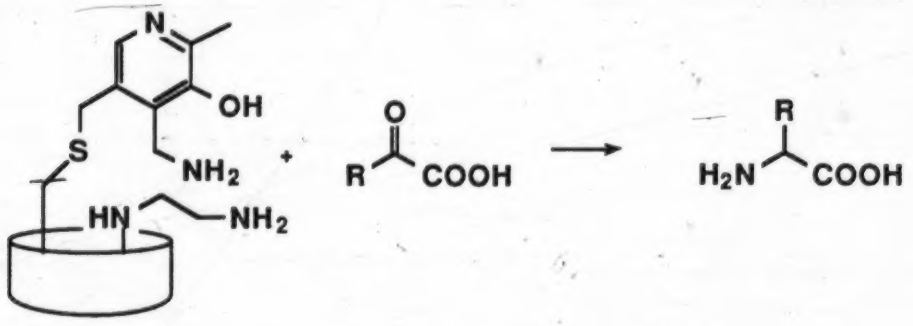
## A. Some Recent Methods for Amino Acid Synthesis

1.) Optically active allylic selenides undergo oxidative [2,3] sigmatropic

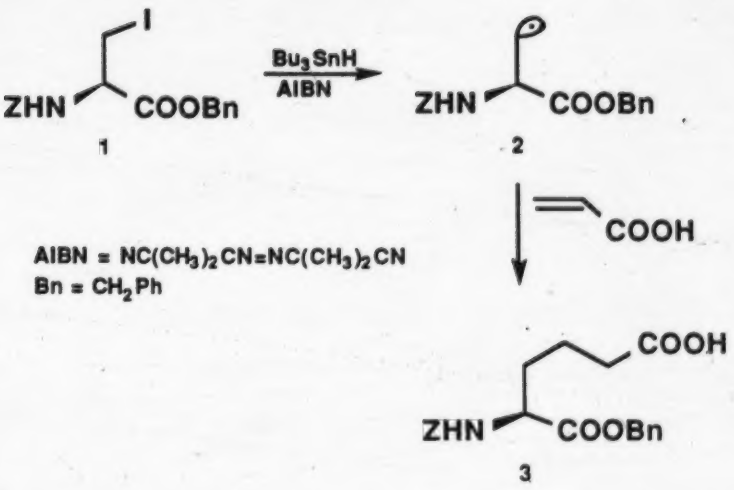


rearrangement to afford, after ozonolysis of the double bond and oxidation of the resulting aldehyde, N-protected optically active α-amino acids in good yield with 78-84% enantiomeric excess (e.e.).[13]

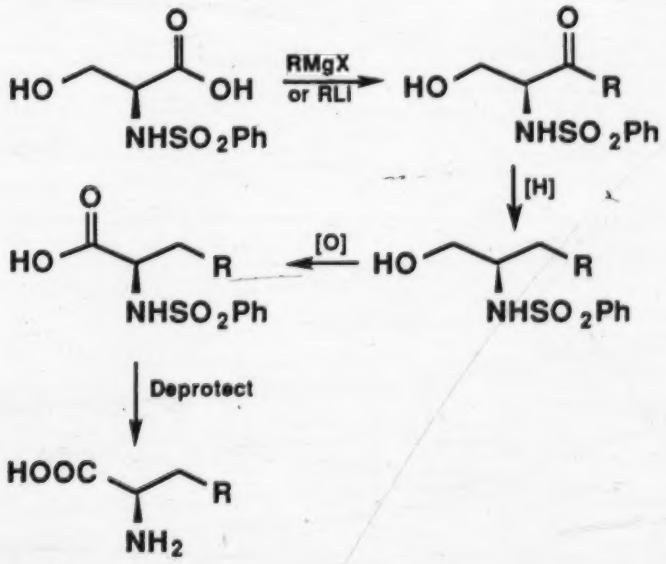
2.) The research groups of Breslow and Tabushi have developed vitamin $B_6$ dependent aminotransferase models that can generate some amino acids in ≥ 95% e.e.[14] Flexible side chains carrying amino groups which promote stereospecific proton transfers have been attached by Breslow and workers to pyridoxamine. Cyclodextrin, a cycloamylose which promotes substrate binding has been coupled with pyridoxamine, in the same laboratories. Tabushi's group has regiospecifically introduced into cyclodextrin functional groups capable of performing proton transfers. The culmination of the efforts of these two research groups has provided enzyme mimics which convert keto acids into amino acids.




3.) Baldwin and coworkers have used a radical addition reaction to prepare an α-amino acid.[15] Protected 3-iodo-L-alanine (**1**) was reacted with tributyltin hydride in the presence of azobisisobutyronitrile (AIBN) to generate the optically active alanyl radical **2** which coupled with acrylic acid to produce protected optically pure α-aminoadipic acid **3**. However, only a 30% yield of pure product was obtained.



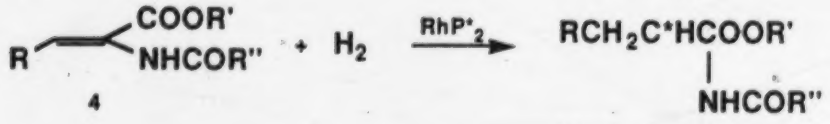

4.) Rapoport has reported a general synthesis of the relatively rare D-α-amino acids from L-serine utilizing as the key step the aminoacylation of organolithium or Grignard reagents.[9] The reaction proceeds with complete retention of stereochemical




integrity if the proper N-protecting group is used. Little or no formation of the tertiary alcohol occurs. Reduction of the ketone to a methylene group followed by oxidation of

the primary alcohol to the acid, and deprotection of the amino group secures the D-α-amino acids in good yield with > 99% e.e.

5.) Under the catalysis of rhodium complexes containing chiral phosphine ligands the asymmetric hydrogenation of some Z-α-acylaminoacrylic acid derivatives 4 has been achieved in optical yields approaching 100% e.e.[16] This approach is equally satisfactory regardless of whether the chirality is at the coordinating phosphorous atom, at a substituent on the phosphorous, at a centre on a chiral backbone linking two phosphorous ligands, or due to the presence of axially dissymmetric bisphosphine ligands.[29,30]



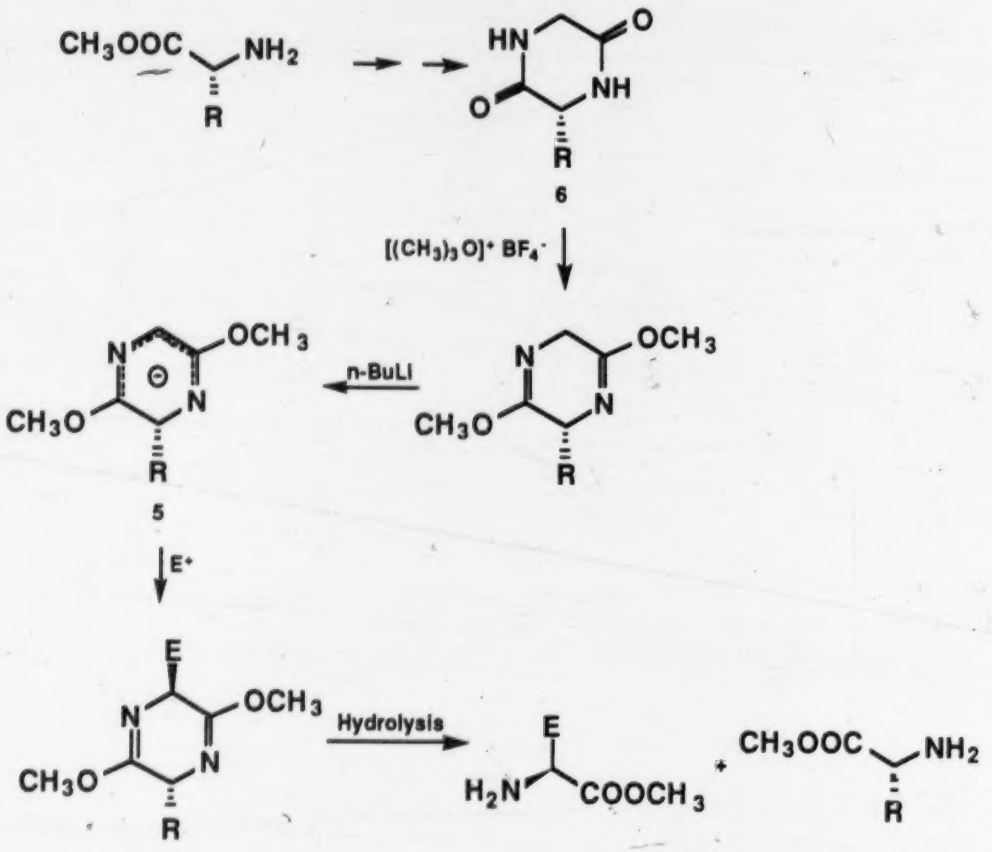

4

The main problem with this route is in the preparation of the unsaturated precursor. When R is aromatic the synthesis of the enamide from the corresponding aldehyde (RCHO) is usually quite efficient, but when it is aliphatic the condensation yields are low.[16]

Mechanistic studies on these hydrogenations have revealed that the stereochemical outcome results from reduction of the catalyst-substrate adduct corresponding to the less favoured binding mode.[31,32] Presumably this is due to its greater reactivity.[31,32]

6.) Alkylations of chiral heterocyclic rings have been used to generate optically active amino acids.[10,11,17] Electrophiles react stereospecifically with enolates of bis-lactim ethers 5 (derived from 2,5-diketopiperazines 6) from the less hindered side of the chiral ring (> 95% diastereomeric excess (d.e.)). Hydrolysis at the imino ether

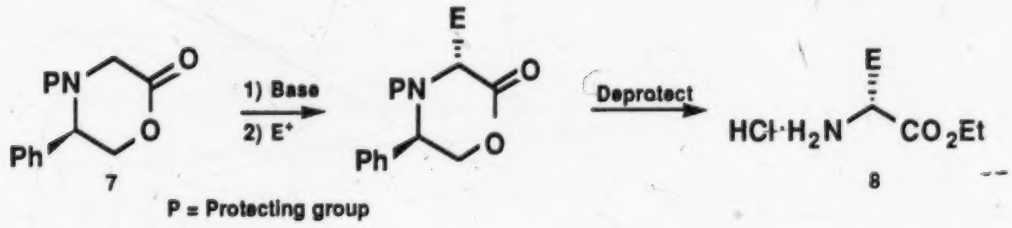
group then affords the optically active α-amino acid methyl esters and regenerates the chiral auxiliary.



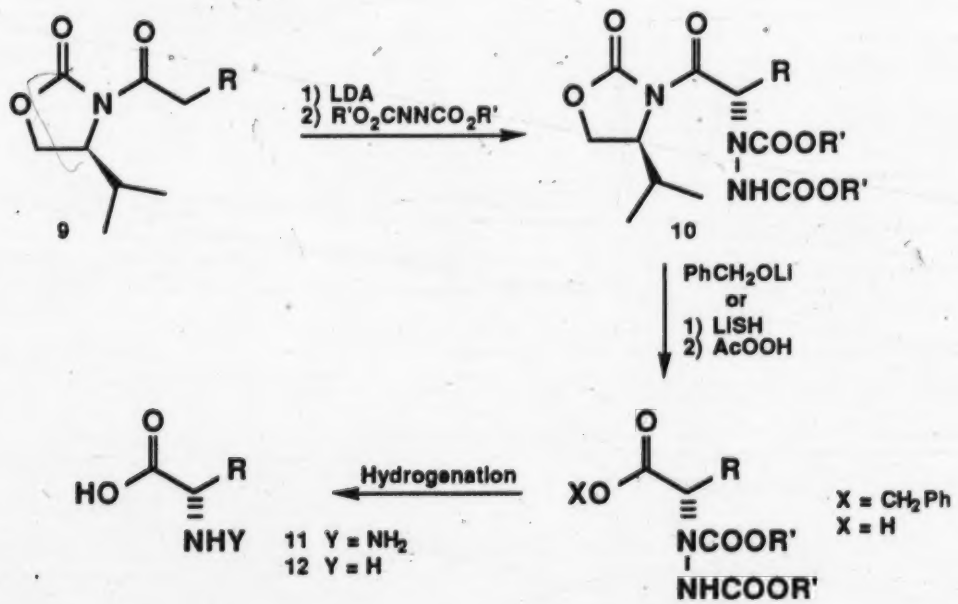

This method allows the synthesis of D or L amino acids from the appropriate antipode of the chiral auxiliary. The electrophiles which have been utilized are alkyl halides, aldehydes, ketones, thioketones, and more recently α, β-unsaturated ketones and esters.[11,12,17,18] This route allows hydrogenation-sensitive moieties to be present and gives very predictable results for a wide range of optically active amino acids, in contrast to the enantioselective hydrogenation of N-acyldehydroamino acids. The latter pathway cannot have functional groups present which are sensitive to hydrogenation,

and suffers from the necessity of conducting preliminary studies in each case to find suitable catalysts and reduction conditions.

7.) Recent results using oxazines and oxazolidinones as chiral auxiliaries provide α-amino acids in high yields and good optical purity. Evans and coworkers have alkylated the enolate of a chiral N-protected oxazinone **7** which after deprotection affords the amino acid esters **8** with ≥ 99% e.e.[19]

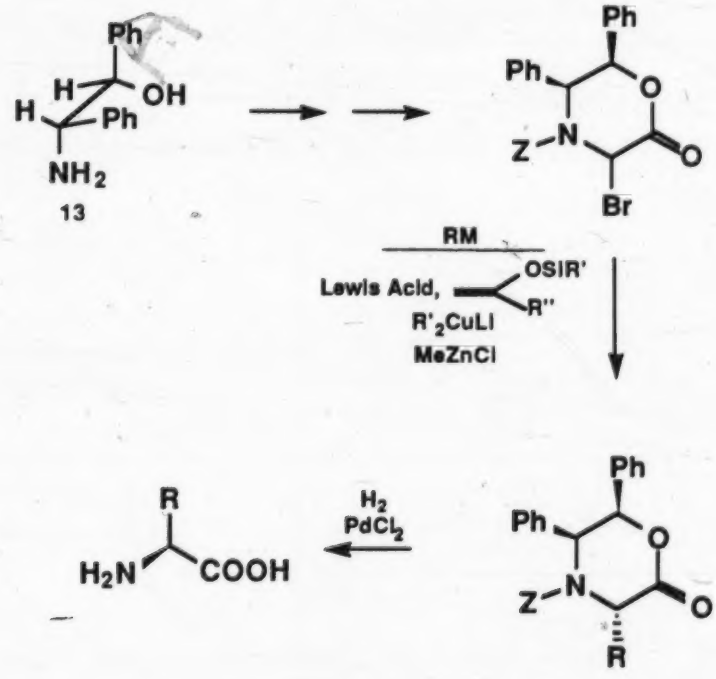



In our laboratories chiral enolates of N-acyl oxazolidinones **9** have been treated with dialkyl azodicarboxylates to give aminated derivatives **10**. These can be deprotected with lithium benzyl alkoxide or lithium hydrosulfide, followed by

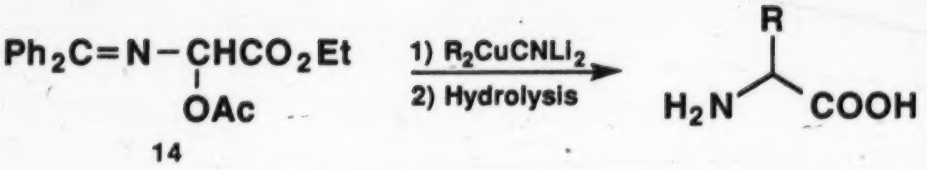


hydrogenation to afford α-hydrazino acids **11** or α-amino acids **12** in good yields with ≥ 90% e.e.[20]

8.) Existing enolate-based methodologies are complemented by new approaches using electrophilic glycinates. For example, D-erythro-α,β-diphenyl-β-hydroxyethylamine (**13**) have been converted to chiral oxazinones which have condensed with silyl enol ethers or organometallic reagents to give α-amino acids in yields ranging from 28 - 71% with 97 - 100% e.e.[21] The yields are better using the silyl enol ethers than the organometallic reagents.




A cationic glycine equivalent, iminium acetate **14**, has been attacked with organocuprates.[33] Fair to good yields (46 - 71%) of DL-α-amino acids were produced.

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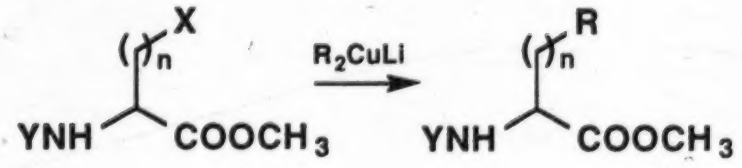
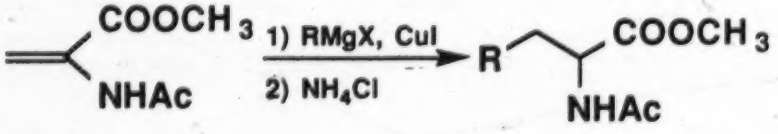
9.) Organocuprates have been used to affect $S_N2$ type displacements on protected amino acids substituted with a halogen or tosyl groups in the β or γ position.




Y=Boc, Ts
X=I, Br, OTs
n=1, 2

When displacement occurs at the γ position,[22] yields and optical purity are higher than when the leaving group is at the β position because of competing elimination.[23,24]

10.) Organocopper chemistry has also been used to prepare racemic DL-N-acetyl amino acid methyl esters **15** from methyl α-acetamidoacrylate (**16**).[34] Conjugate

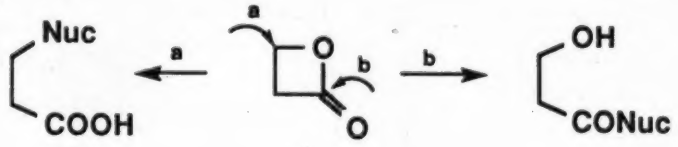



16 15

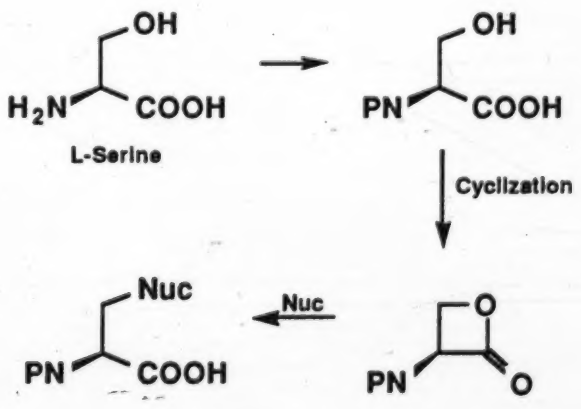
addition of Grignard reagents to **16** in the presence of CuI catalysis affords fair to good yields of α-amino acid derivatives.

## B. Nitrogen-Substituted β-Lactones as Possible Precursors for Amino Acids

Lactones having a four membered ring are attractive synthetic intermediates because ring strain (22.7 kcal/mole)[35] encourages nucleophilic attack at either the β-carbon (path a) or carbonyl (path b).

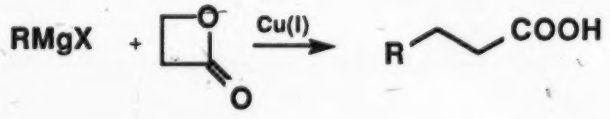



Hard nucleophiles such as hydroxide, alkoxide, most organolithiums, and Grignard reagents attack at the carbonyl whereas soft nucleophiles like halides, acetate, thiolates, selenides and organocuprates attack at the β-methylene carbon.[36-55] Based on these results it appeared that optically active N-protected α-amino-β-lactones could in principle be regiospecifically ring opened to produce optically active N-protected amino acids. Since the chiral centre would not be directly involved in the reaction, optically pure products should be obtained. The readily available amino acid serine could serve as a starting material for the required N-protected α-amino-β-lactones.



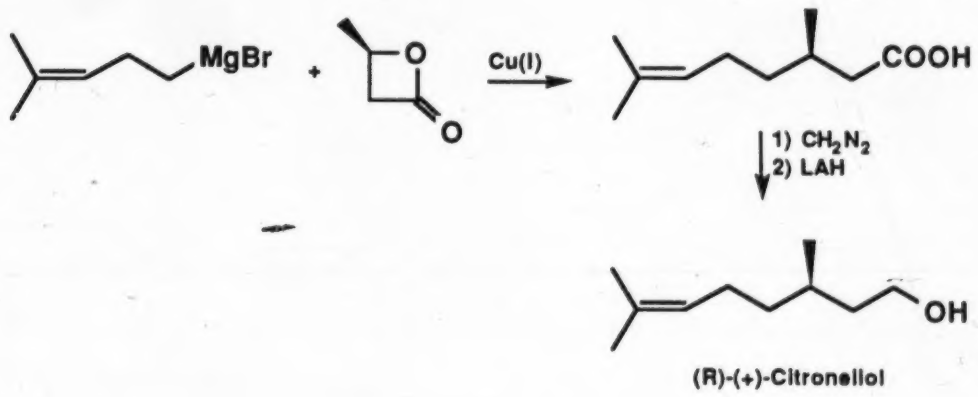
The first report of ring opening of a β-lactone with an organometallic reagent was by Gresham *et al.*[36] They found phenylmagnesium bromide attacks β-propiolactone at the acyl carbon whereas benzylmagnesium chloride attacks at the β-methylene carbon. Polymerization and attack of halide to give β-haloacids were side reactions which were much more prevalent when the halide was bromide. Subsequently, Stuckwisch and Bailey[37] reported that benzyllithium, diphenylzinc, allylmagnesium bromide, and organocadmium compounds attack β-propiolactone at the β-carbon. In the case of the organocadmiums, thermal stability and steric bulk were the limiting factors in producing good yields of ring-opened products. In contrast, phenyllithium and *o*-anisyllithium attack at the acyl carbon of β-propiolactone to give ketones and/or alcohols.[37]

Normant and coworkers[38] have employed Cu(I) catalysis of Grignard reagent addition to obtain regiospecific opening of such lactones at the β-carbon to form long chain carboxylic acids.

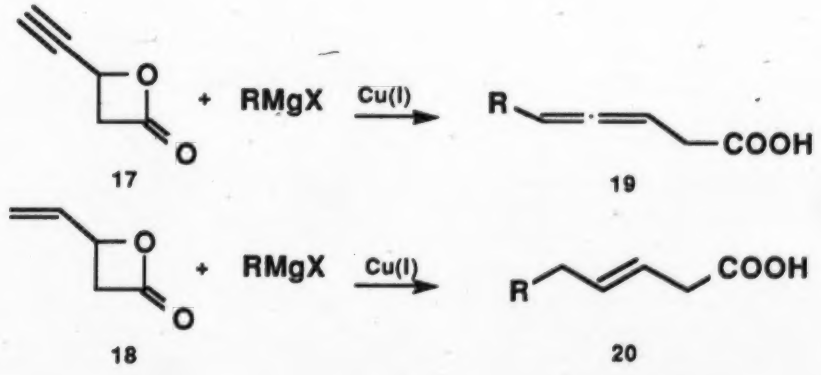



The yields with vinyl and allylic systems are improved when stoichiometric organocuprates are employed. During the course of their work it was also noted that mixed heterocuprates transfer a substantial amount of the heteroanion. This problem was overcome by using mixed alkynyl cuprates. Fujisawa and coworkers have independently observed ring opening of β-lactones with Grignard reagents with Cu(I) catalysis[39] and have extended the results to classical Gilman reagents and halomagnesium cuprates.[40] The halomagnesium cuprates give only slightly better yields with the exception of the phenyl cuprate where yields are dramatically increased

from 20% to 80%. Not surprisingly, substitution of the β-lactone decreases the yields, presumably due to increased steric hindrance.[39,40] This research group exploited the regiospecificity of organocuprate attack to synthesize various naturally occurring compounds including perfume constituents,[41,42] various terpenes,[43-45] insect pheromones,[46,47, 56] and antibiotics.[57,58]

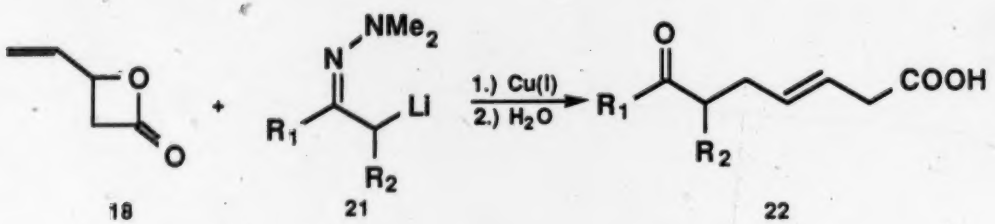



Interestingly, β-ethynyl **17**[57,59] and vinyl β-lactones **18**,[56,58,60-62] provide allenic **19** and 3-alkenoic acids **20** via $S_N2'$ ring cleavage.




Lithium enolates could be expected to attack β-lactones at the carbonyl. Condensation of the lithium enolate of acetone with β-vinyl-β-propiolactone **18** gave

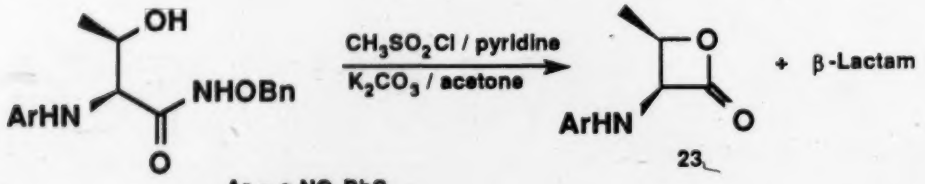
neither $S_N2$ attack at the β-carbon nor $S_N2'$ at the terminal vinyl carbon.[60] The copper enolate only gave 21% of the desired acids. However, the copper-catalyzed reaction of the corresponding N,N-dimethylhydrazone **21** enolates produced the desired 7-oxo-(E)-3-alkenoic acids **22** in good regio and enantioselective yields via the $S_N2'$ attack at the terminal vinyl carbon.[60]




18 21 22

These results indicated that β-lactone cleavage with new carbon-carbon bond formation was possible.

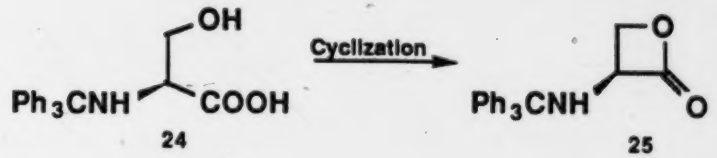
Previous syntheses of N-protected α-amino-β-lactones have relied either on carboxy group activation of β-hydroxy amino acid derivatives or on generation of a leaving group at the β-position in such compounds. In an attempt to prepare



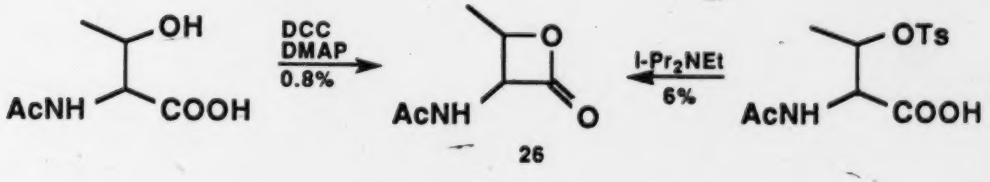

23

N-(*o*-nitrophenylsulfenyl) threonine β-lactam, Gordon *et al* [63] produced the β-lactone **23** as a side product via carboxy group activation. Sheehan and coworkers[64] employed diisopropylcarbodiimide to cyclize N-trityl-L-serine (**24**) to the corresponding β-

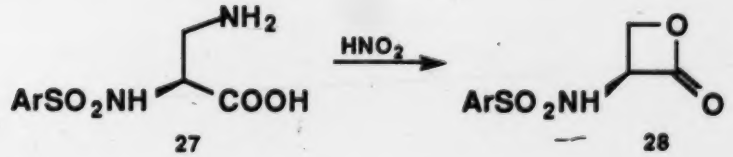
lactone **25** in 15% yield. This procedure was later improved (26% yield) by Shanzer and Libman[65] who added 4-dimethylaminopyridine (DMAP) as an acylation catalyst.[66]



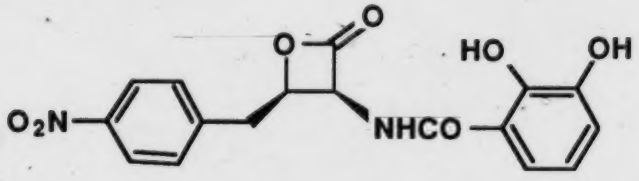

Using the same type of reaction, Parker *et al*[67] obtained a 0.8% yield of N-acetyl threonine β-lactone **26**. They isolated this compound from Bacillus species SC 11,480. An antimicrobial evaluation of racemic **26** showed it has very weak antimicrobial activity.[68] An alternative approach involving generation of the tosylate at the β-position followed by cyclization of the β-lactone with Hünigs base gave only 6% of the desired compound.




Intramolecular displacement of nitrogen from the diazonium salt of 2-arylsulfonyl-3-aminopropionic acids **27** affords up to 62% yields of the β-lactones **28**[69,70] but this reaction is limited to N-arylsulfonyl derivatives which are more difficult to deprotect.



Mitsunobu reaction of β-hydroxy carboxylic acids can produce β-lactones and/or alkenes depending on reaction conditions.[71-73] Under standard Mitsunobu conditions[74] (triphenylphoshine, diethyl azodicarboxylate and tetrahydrofuran (THF)) Parker *et al* [67] prepared N-phenylacetyl serine β-lactone in 1.4% yield. Recently in our laboratory it was found that the Mitsunobu reaction can afford high cyclization yields of N-protected serine derivatives if these derivatives are added to a preformed adduct of triphenylphosphine and dimethyl azodicarboxylate at low temperatures.[48] Using this methodology β-substituted alanines were produced by attack with halide or heteroatom based nucleophiles. This method may also prove useful in the synthesis of β-lactone containing natural products some of which have been shown to exhibit biological activity.[67,68,75-81] For example, obafluorin is an antibiotic isolated from *Pseudomonas fluorescens*.[75,76]



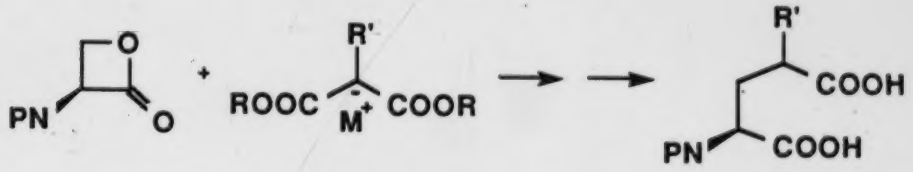

Obafluorin

. In addition, α-amino β-lactones have been used to produce polymers.[69] Certain β-lactone derived polymers are known to be suitable for producing polymers which are suitable for fibers and thermoplastics.[82]

Success in forming N-protected serine β-lactones by Mitsunobu reactions and in opening these compounds with heteroatom nucleophiles[48] led us to investigate opening with carbon nucleophiles. The ample precedent of regiospecific ring opening of β-lactones with organocuprates provided by the research groups of Fujisawa and Normant suggested these soft nucleophiles would be suitable to attack N-protected

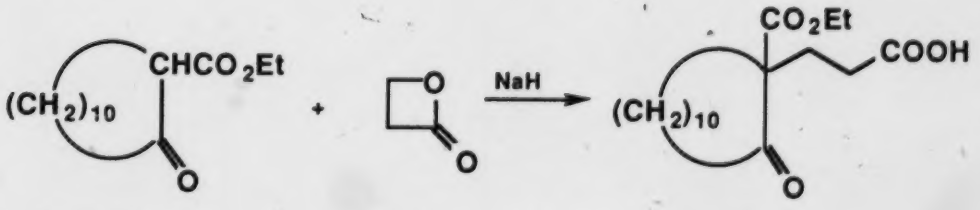
serine β-lactones. Organocuprate attack on these β-lactones would provide a new route to a vast array of α-amino acids

Malonates are also soft carbon nucleophiles and as such, should cleave the β-lactones at the β-carbon. Hydrolysis of the resulting esters with concomitant decarboxylation would give γ-substituted derivatives of glutamic acid **29**. Malonates and other active methylene compounds have opened



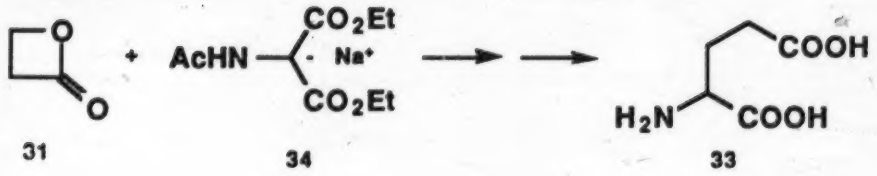

**29** R' = H

β-lactones.[49-51] For example compound **30**, a useful perfume intermediate was prepared from β-propiolactone (**31**)and 2-ethoxycarbonylcyclododecanone (**32**) in 57% yield.[83]



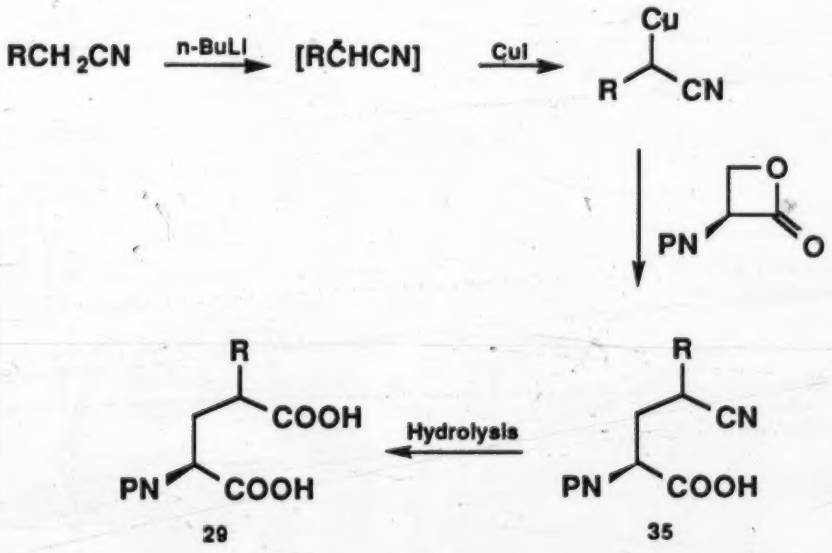

**32** **31** **30**

Similarly DL-glutamic acid (**33**) has been prepared from β-propiolactone (**31**) and the sodium salt of diethyl acetamidomalonate (**34**) in 87% yield.[84]



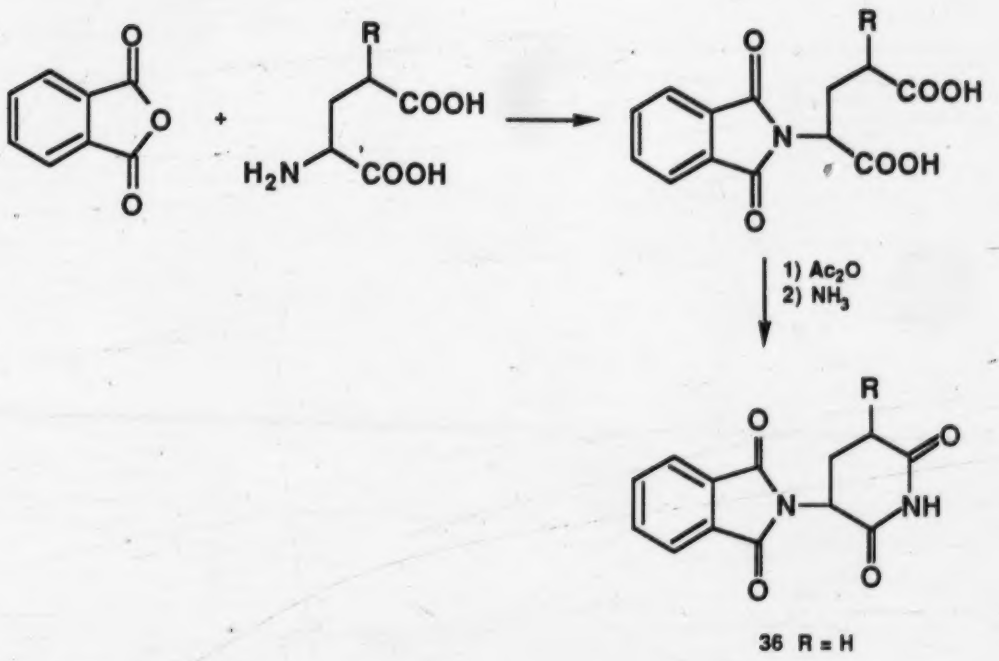

31 34 33

Another potential route to substituted glutamates relies on observations of Corey and Kuwajima.[85] Cyanomethylcopper, simply prepared by adding one equivalent of *n*-butyllithium to acetonitrile followed by one equivalent of cuprous iodide, affects $S_N2$ type displacements of allylic bromides. Extension of these results to primary nitriles may prove useful in preparing γ-substituted derivatives of glutamic acid **29** by regiospecific ring opening of N-protected α-amino β-lactones followed by hydrolysis of the nitrile **33** to a carboxylic acid.




29 35

Interest in γ-substituted derivatives of glutamic acid **29** stems from their potential to serve as precursors in the synthesis of substituted derivatives of thalidomide (**36**). The substituent allows the attachment of a probe onto the teratogenic thalidomide




36 R = H

moiety so that its path from the mother through to the fetus can be monitored. Such probes could include photoaffinity labels, fluorescent moieties, spin labels, and functional groups which would be attached to other "recognition molecules" or to solid supports. Results from such experiments would help to elucidate which cell constituents are involved in the manifestation of thalidomide's dysmorphogenic effects.

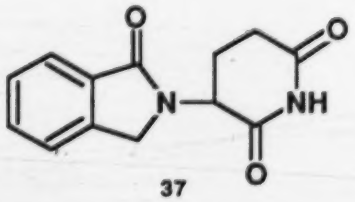
## C. Thalidomide and its Background

Thalidomide (**36**)was first prepared in 1956 at Chemie Grünenthal in West Germany[86] and was promoted as being a very safe and effective sedative-hypnotic.[86,87] In contrast to many other sedatives it does not decrease muscular coordination nor affect blood pressure, heart rate, pulmonary function or urine output[87,88] at a dose of 50-200 mg per day orally. Numerous suicide attempts with doses up to 14.4 g resulted only in sleep induction with no other observable side effects.[89] The apparent safety of this drug coupled with its high therapeutic index led

to its widespread use. Early in 1960 it became a commonly prescribed antiemetic for pregnant women in several countries, but by late 1961 it was linked to an increase in the number of congenital birth defects.[90-92]

The range of defects varied from minor thumb anomalies to the total absence of limbs.[93] Although skeletal defects were the most common, gastrointestinal, genito-urinary, ocular, auditory, cardiovascular and respiratory defects were observed. Ultimately approximately 10,000 children were affected.[93] It is not surprising the early mortality rate among these children was 45%.[94] The specific time during pregnancy when the drug is taken is critical with defects appearing only if the drug is ingested from day 34 to 50 since the last menstrual period.[94] Which organ was affected varied depending on the time of intake of the drug and correlated well to the sequence of organogenesis. Rats and mice are relatively resistant, rabbits are sensitive, and monkeys and humans very sensitive to the teratogenic effects of thalidomide.[94]

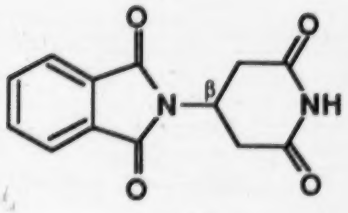
Structure activity relationships have yielded some insight but allow only a few of the multitude of suggested theories to be discarded.[93-97] An intact phthalimide moiety was thought to be indispensible but partial reduction to phthalimidine **37** gives a



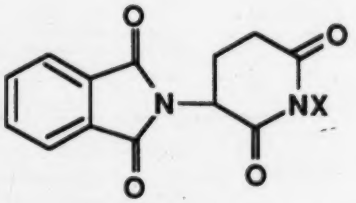

**37**

compound which is at least three times as teratogenic as thalidomide.[95,96] This indicates alteration of the phthalimide ring is permissable without loss of teratogenic activity. The iso-thalidomide or the β-form **38** is not teratogenic.[95] Replacement of the glutarimide ring with a succinimide or phenyl ring resulted in a loss of activity.[95]

Methylation of the glutarimide ring nitrogen decreases activity. N-methoxy (**39**) and N-hydroxy thalidomide (**40**) are also inactive as teratogens. Replacement of the

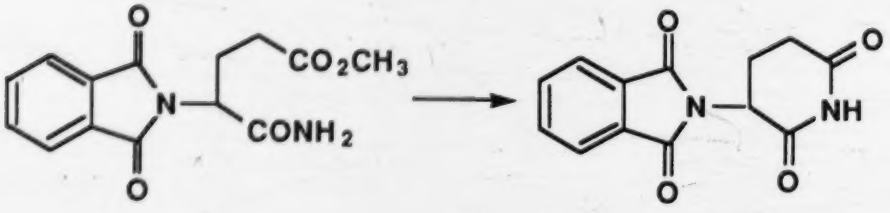



**38**



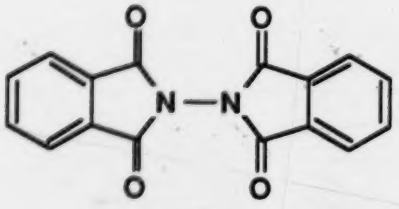

**39** X = $OCH_3$
**40** X = OH

glutarimide ring with a glutarimide ester **41** retains activity, presumably because of its conversion to thalidomide *in vivo*.[95]



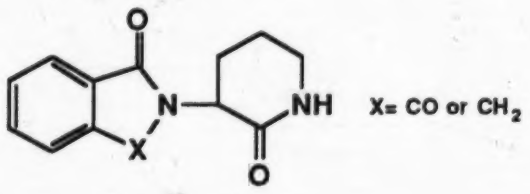

**41** **36**

It was once thought the glutarimide ring was essential for teratogenic activity but phthalimido-phthalimide (**42**) was shown to be at least as teratogenic as thalidomide.[96]




**42**

Recently it was demonstated the glutarimide ring can also be reduced to an oxopiperidine **43** with retention of teratogenic activity.[97]



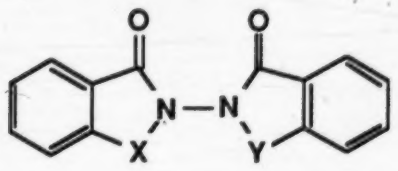

**43**

A number of theories have been advanced to explain the teratogenic activity of thalidomide[93-98] some of which are mentioned here. The trapping theory postulates that thalidomide readily penetrates the placental barrier where it can be hydrolyzed. The resulting polar products are thus trapped inside the fetus where their concentration builds to levels which could be capable of producing teratogenicity. It was suggested that thalidomide, due to the presence of the glutarimide moiety, might interfere with glutamic acid metabolism. Another theory postulates that thalidomide's teratogenic effects result from its ability to act as an acylating agent. More recently it has been proposed by Gordon *et al*[98] that oxidative metabolism to an arene oxide occurs, and that this metabolite then reacts with biologically important macromolecules to produce dysmorphogenic effects. Using their assay these authors have shown that addition of epoxide hydrolase abolishes thalidomide activity while addition of inhibitors of this enzyme increase activity. The presence of $^3H_2O$ in the plasma of rabbits and the recovery of phenolic metabolites from their urine after administration of aromatically tritiated thalidomide provide further evidence for an arene oxide intermediate. Another theory proposes teratogenicity arises through the ability of these drugs to suppress immune rejection by the mother of a spontaneously deformed fetus. However the

increased number of resorptions seen in treated animals makes the validity of this theory questionable.[94]

Other observations implicating the immunosuppresive properties of thalidomide have appeared to account for thalidomide's efficiency in treating several skin disorders,[99,100] including Behçets syndrome,[101,102] Weber-Christian disease,[103] several forms of lupus erythematosus,[104-107] orogenital ulceration[108] and leprosy.[109-111]

The observation of teratogenic activity in N-phthalimidophthalimide (**42**) led us to question if the analogs with one or both imides converted to amides would be teratogenic. The acylation theory would predict **44** with an imide moiety still present would be teratogenic and **45** devoid of any labile acylation site would not be. The arene oxide theory would predict that in principle, both could be teratogens.



**42** X, Y= CO Teratogenic
**44** X= $CH_2$, Y= CO
**45** X, Y= $CH_2$

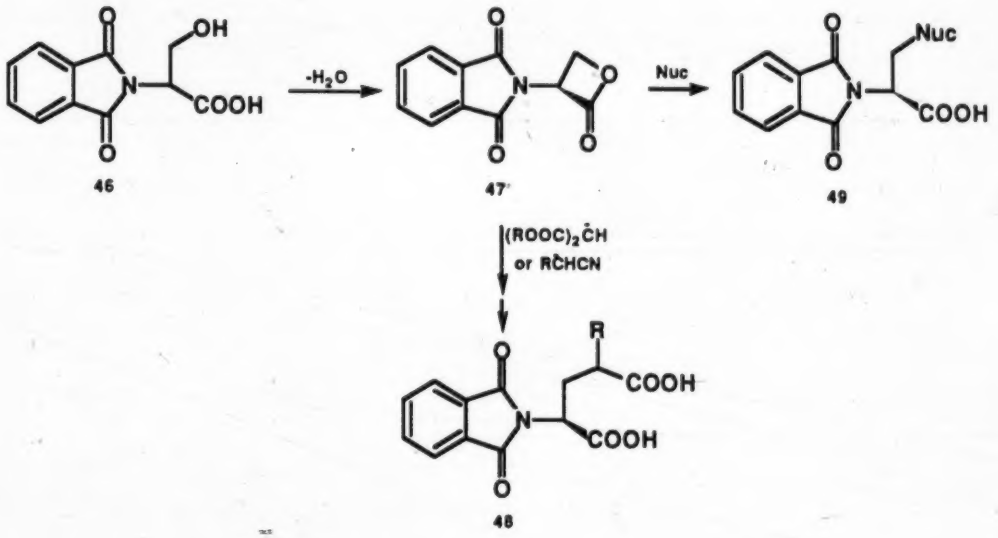
A review of the literature revealed the analog **45** had been prepared but not biologically tested.[112] The other analog **44** was unknown. Testing of these compounds would improve our understanding of structure activity relationships involved in thalidomide teratogenicity. These results would complement those obtained from γ-substituted thalidomides. They might also serve in the search for nonteratogenic analogues of thalidomide which would be useful in the treatment of leprosy and other disorders.

This thesis describes the preparation of thalidomide (**36**), N-phthalimidinoglutarimide (**37**) and N-phthalimidophthalimide (**42**) N-

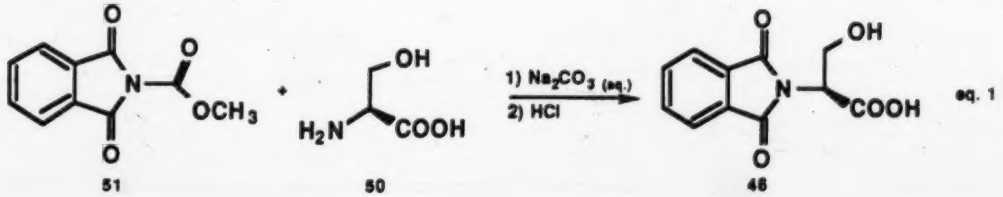
phthalimidophthalimidine (**44**), N-phthalimidinophthalimidine (**45**). In addition, the synthesis of several α-amino-β-lactones and their reactions with both heteroatom and carbon nucleophiles to afford α-amino acid derivatives was investigated. The goal of the latter work was to determine the potential of this approach for the production of substituted amino acids which could be eventually converted to thalidomide analogs.

## RESULTS AND DISCUSSION

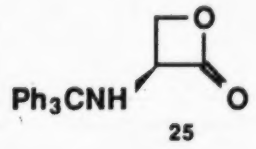
Retrosynthetically it can be seen N-phthaloyl-L-serine (**46**) can serve as a precursor to N-phthaloyl-L-serine-β-lactone (**47**). This β-lactone could then be opened by malonates or α-anions of nitriles to furnish γ-substituted glutamic acid derivatives **48** or with other nucleophiles to afford β-substituted alanines **49**.




Reaction of L-serine (**50**) with N-carbomethoxyphthalimide (**51**)[113] under basic aqueous conditions affords **46** in an unoptimized 29% yield after acidic workup (equation 1).



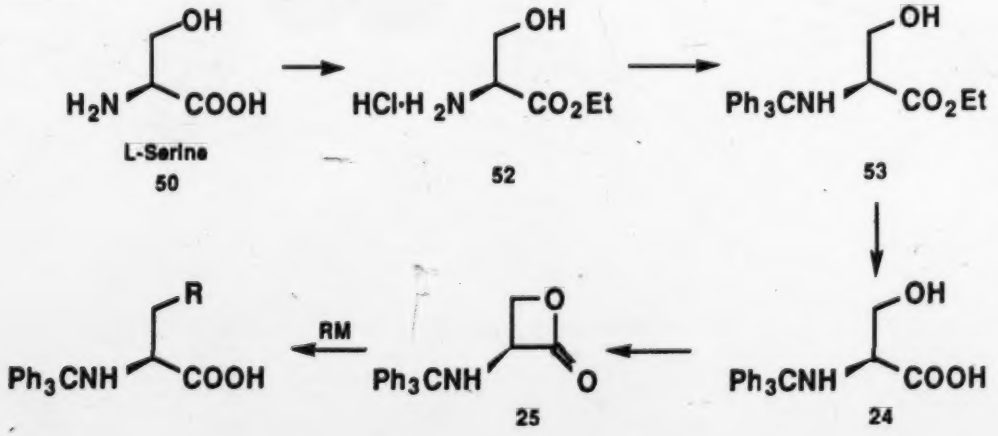
N-Phthaloyl-L-serine (**46**) could be cyclized to the β-lactone **47** under Mitsunobu conditions (triphenylphosphine, diethyl azodicarboxylate) as evidenced by appearance of an absorption in the infrared (IR) spectrum at 1845 $cm^{-1}$ during the course of the reaction.[114] However, the β-lactone was unstable and could not be purified by either recrystallization or chromatography, presumably because of the reactivity of the phthalimide moiety. Reaction with sodium dibenzylmalonate *in situ* failed as well. Attention then shifted to preparing N-trityl-L-serine-β-lactone **25** because the trityl group should be easily removed by acid at a later stage.[115]




25

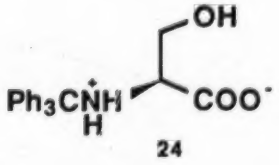
Shanzer and Libman[65] had prepared this β-lactone by modifying the procedure of Sheehan *et al*[64] (Scheme 1). They increased the yield from 15% to 26% by adding a catalytic amount of dimethylaminopyridine (DMAP).[66] Presumably to facilitate purification, both groups used diisopropylcarbodiimide as the cyclization reagent rather than the more common dicyclohexylcarbodiimide.

Scheme 1



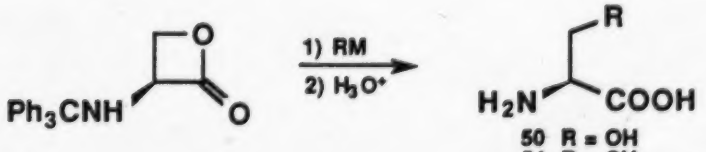
L-Serine ethyl ester hydrochloride (**52**) was prepared from L-serine (**50**) using anhydrous ethanol and HCl gas.[116] This salt was converted to the free amine with triethylamine and protected with trityl chloride to provide the ester **53** in 92% yield.[117] Saponification with alcoholic potassium hydroxide, followed by careful acidification to pH 4 with phosphoric acid gave N-trityl-L-serine (**24**) in 58% yield.[118] If the reaction medium becomes too acidic loss of the acid labile trityl group occurs.

Cyclization to the β-lactone **25** with diisopropylcarbodimide, catalyzed by DMAP, followed by flash chromatography with toluene rendered 10% of N-Trityl-L-serine-β-lactone (**25**). Attempts to cyclize **24** under Mitsunobu conditions (triphenylphosphine, diethyl azodicarboxylate, THF, -78°C) failed, presumably because of the zwitterionic character of this compound.[114]



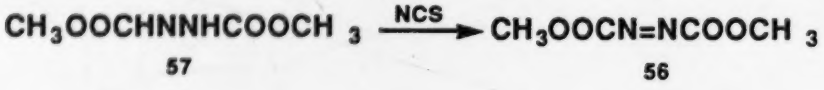

**24**

Reaction of β-lactone **25** with dimethyl copper lithium and acidic workup gave a complex mixture of products which contained serine (**50**) and α-aminobutyric acid (**54**). The low yields of cyclization of trityl-serine **24** and of its nucleophilic opening encouraged use of another protecting group.



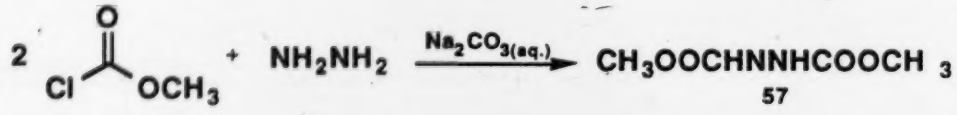

**50** R = OH
**54** R = $CH_3$

N-Benzyloxycarbonyl-L-serine-β-lactone (**55**) was prepared using modified Mitsunobu conditions.[48] The use of dimethyl azodicarboxylate (**56**) rather than the more conventional diethyl azodicarboxylate is preferred for ease of product purification. Diethyl hydrazodicarboxylate has a $R_f$ value very close to that of the desired β-lactone,whereas dimethyl hydrazodicarboxylate (**57**) and the desired β-lactone **55** have a large enough difference in their $R_f$ such that purification by column chromatography is feasible. Dimethyl azodicarboxylate (**56**) was prepared[119] from dimethyl hydrazodicarboxylate (**57**) by



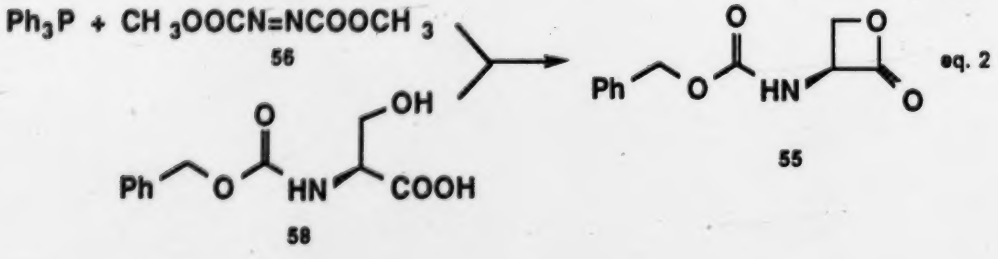

**57** **56**

oxidation with N-chlorosuccinimide (NCS).[120] Reaction of methyl chloroformate with hydrazine hydrate gave the hydrazodicarboxylate **57**.[121]

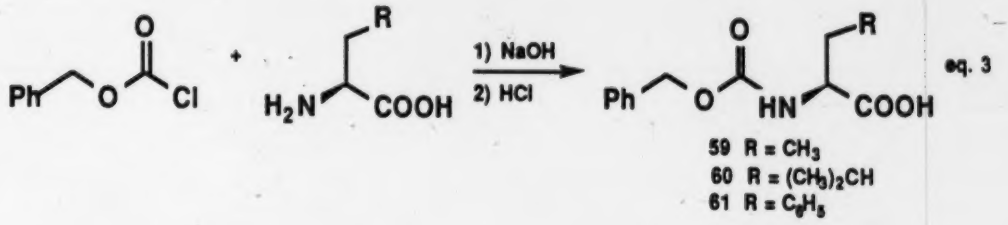



**57**

Addition of N-benzyloxycarbonyl-L-serine (**58**) to a preformed betaine adduct of triphenylphosphine and dimethyl azodicarboxylate in THF at -22°C furnished the β-lactone **55** (equation 2).

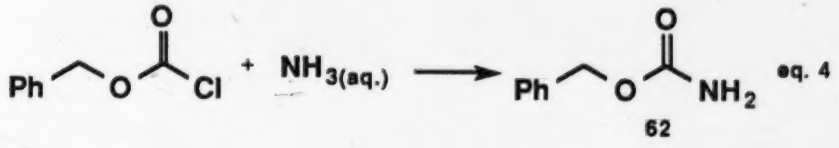


In order to aid identification of products resulting from condensation of the β-lactone **55** with organocuprates, authentic samples of the desired N-protected amino




**59** R = $CH_3$
**60** R = $(CH_3)_2CH$
**61** R = $C_6H_5$

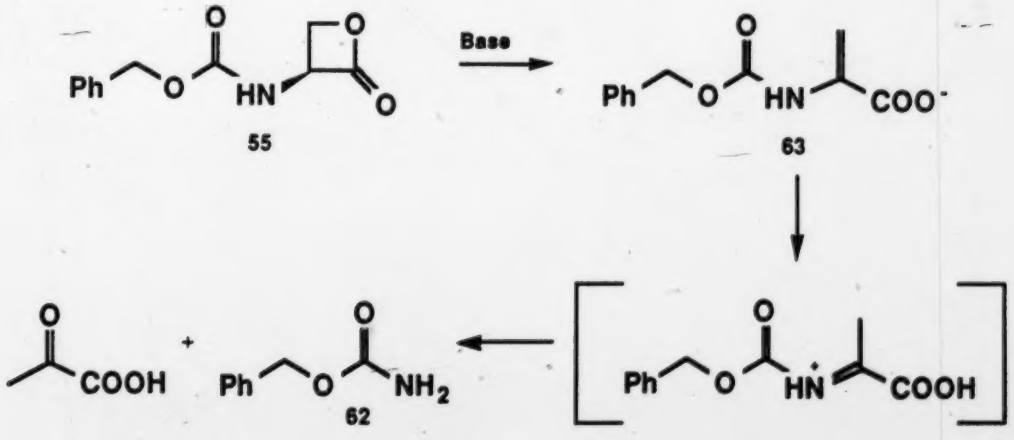
acids **59-61** were prepared by reaction of benzyl chloroformate with the corresponding amino acids under Schotten-Bauman conditions[116] (equation 3). An authentic sample of a potential side product, benzyl carbamate (**62**) was prepared in the same manner (equation 4).




**62**

Previous work had shown that base can cause cleavage of β-lactones to olefins.[49,114,122] In the case of α-amino-β-lactones, dehydroamino acid derivatives

Scheme 2



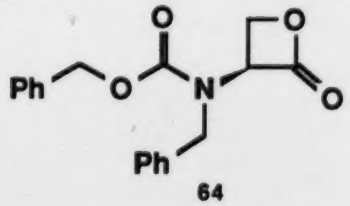

**55** **63** **62**

**63** are the products of such cleavage. Hydrolysis of these labile entities occurs in aqueous acid. Benzyl carbamate (**62**) could arise by eliminative β-lactone ring opening followed by hydrolysis of the resulting unsaturated protected amino acid (Scheme 2).

Reaction of β-lactone **55** with classical Gilman reagents (prepared from purified CuI[123] or $CuBr \cdot Me_2S$,[124]) and higher order Lipshutz reagents [125] failed to give good yields of attack at the β-methylene carbon.

Boron trifluoride-etherate promotes the nucleophilic addition of organolithiums to oxiranes and oxetanes.[126] The presence of $BF_3 \cdot Et_2O$ in reactions of higher order Lipshutz reagents with oxiranes and α,β-unsaturated ketones has been shown in some cases to lead to dramatic enhancements in reaction rate and/or product yields relative to those in the absence of this Lewis acid.[127] However, catalysis of methyllithium addition and higher order Lipshutz reagent addition by $BF_3 \cdot Et_2O$ failed to secure the desired acids in synthetically useful yields.

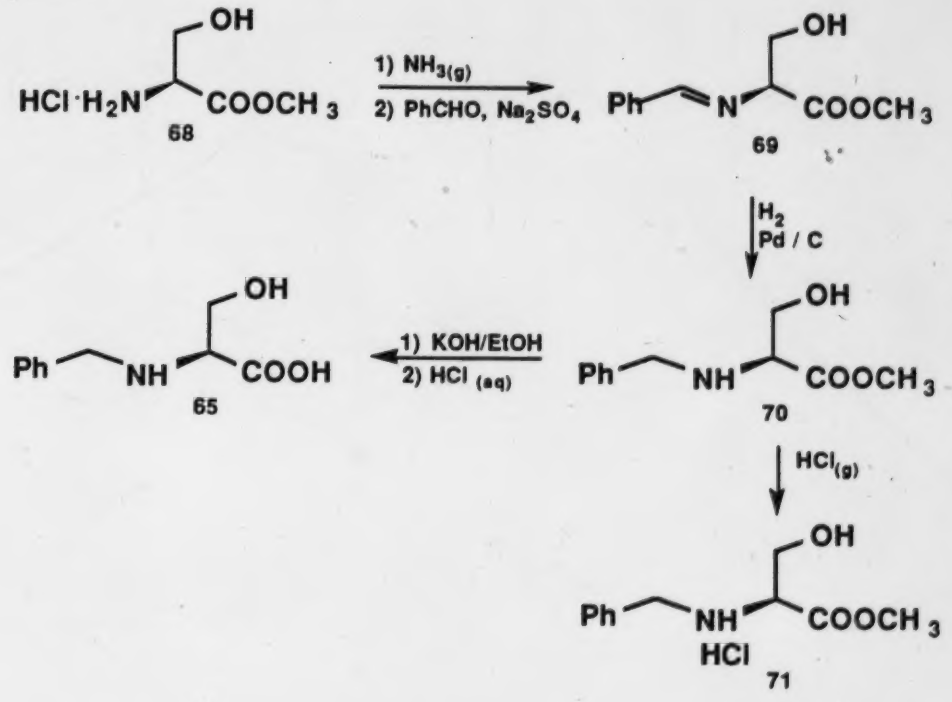
Since the pKa of the carbamate hydrogen of β-lactone **55** is approximately 16, it is conceivable that the organometallic reagent abstracts this hydrogen to form a monoanion which is then less susceptible to attack by another equivalent of organometallic species. This monoanion may attack another molecule of β-lactone to initiate a polymerization process. To circumvent these problems a β-lactone with a fully substituted nitrogen was desireable. A N,N-diprotected β-lactone **64**, where both groups could be removed in one step, was prepared from N-benzyl-L-serine (**65**). The



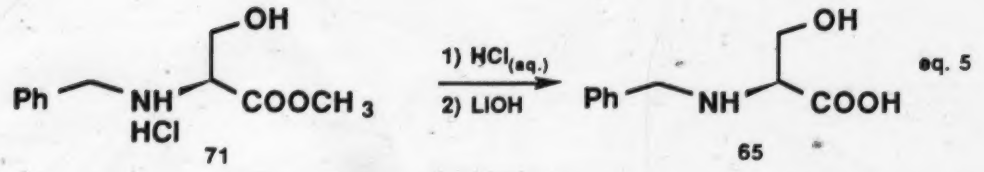

**64**

D-isomer **66** was prepared from N-benzyl-D-serine (**67**). Initially N-benzyl-L-serine (**65**) was prepared by reaction of L-serine methyl ester hydrochloride (**68**) with benzaldehyde in the presence of a drying agent, $Na_2SO_4$, reduction of the resulting imine **69** with hydrogen[128] at 20 psi in the presence of 5% Pd/C catalyst and saponification of the ester **70** with alcoholic potassium hydroxide [129] (Scheme 3).

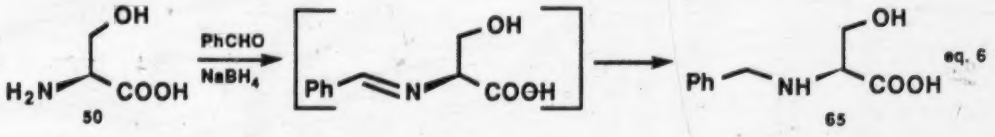
Scheme 3



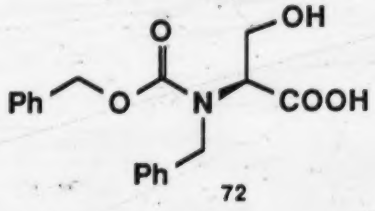

The N-benzyl-L-serine methyl ester (**70**) could be stored as its hydrochloride salt **71**. This could be readily obtained by adding HCl gas to an ether solution of the methyl ester.[128] The N-benzyl-L-serine (**65**) could be prepared in 65% yield by refluxing the methyl ester hydrochloride **71** with aqueous hydrochloric acid for one hour and then adjusting the pH to 5 with lithium hydroxide [128](equation 5).



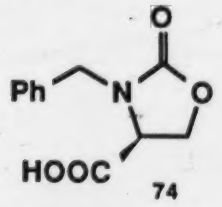

A more efficient route to the N-benzyl-L-serine involved reaction of L-serine (**50**) with distilled benzaldehyde in the presence of sodium borohydride[130] to give stereochemically pure N-benzyl-L-serine (**65**) in 36% yield after recrystallization (equation 6). N-Benzyl-D-serine (**67**) was prepared similarly.




Treatment of N-benzyl-L-serine (**65**) with benzyl chloroformate under mild basic conditions [131] provided **72** in 47% yield. The D-isomer **73** was prepared similarly from N-benzyl-D-serine (**67**). Attempts to increase the yield of N,N-diprotected serine

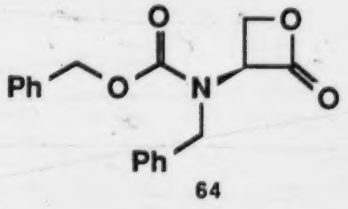



**72** with longer reaction times or heating caused formation of an oxazolidone ring **74** by intramolecular transesterification. The hydroxyl group of the desired product attacks the carbamate carbonyl with concomitant five membered ring formation and elimination of benzyl alcohol.




74

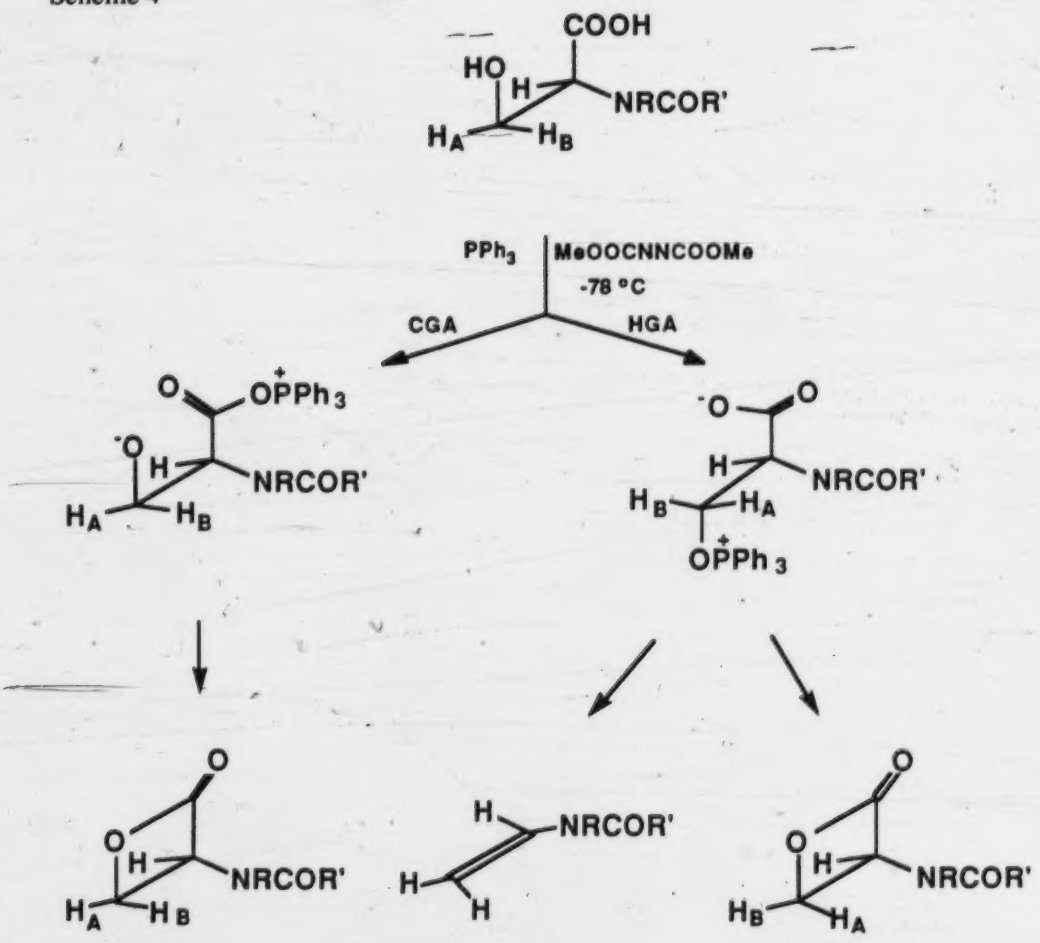
Cyclization of the β-hydroxy acid **72** under modified Mitsunobu conditions (triphenylphosphine, dimethyl azodicarboxylate, THF, -78°C) afforded N-benzyl-N-benzyloxycarbonyl-serine-β-lactone **64** in 71% yield.




64

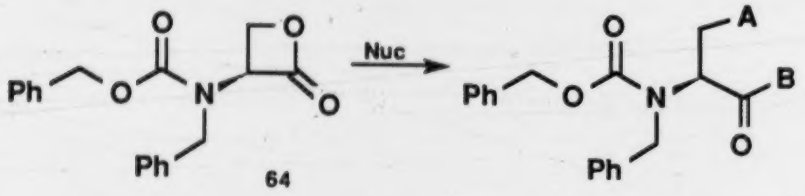
Subsequent to preparation of the monoprotected N-benzyloxycarbonyl-serine-β-lactone **55** it was found[48] that adding the β-hydroxy acid to the preformed adduct of triphenylphosphine and dimethyl azodicarboxylate at -78°C rather than -22°C increased the yields of the β-lactones. At higher temperatures decarboxylative elimination becomes the predominant reaction to afford the olefin.[71-73,132] Although the detailed mechanism of formation of **64** has not been studied, it is likely that this conversion is similar to the cyclization of the monoprotected analog **55**.[52] Previous investigations on the general mechanism of the Mitsunobu reaction have shown that an initially-formed triphenylphosphine azodicarboxylate adduct[133,134] can react with β-hydroxy acids to give either hydroxy group activation (HGA) or carboxy group activation (CGA)[71,72] (Scheme 4).

Scheme 4




Although intermolecular Mitsunobu coupling of alcohols with carboxylic acids to form esters generally proceeds by the HGA pathway,[74,133] this type of activation of β-hydroxy acids has been reported to result primarily in decarboxylative elimination to olefinic products.[71-73,132] In certain alkyl-substituted cases β-lactone products arose principally by the CGA pathway,[71,72] but recent work has shown that lactonization of $^{18}O$ and $^{2}H$ labelled N-benzyloxycarbonyl-L-serine (**58**) occurs by the HGA pathway.[52]

We have sucessfully regiospecifically opened β-lactone **64** with a number of oxygen, nitrogen, sulfur and halogen nucleophiles (Table I).




64

TABLE 1. Reaction of N-Benzyl-N-Benzyloxycarbonyl-α-Amino Serine β-Lactone (**64**) with Non-Carbon Nucleophiles.

| Product | Nucleophile | Conditions | A | B | Yield |
|---|---|---|---|---|---|
| **75** | $NH_{3\,(g)}$ excess | THF 0 °C 2h,rt 9h | $NH_3^+$ | $O^-$ | 37 |
| **76** | " | " | OH | $NH_2$ | 51 |
| **77** | KOAc (7.0 eq.) | DMF/$H_2O$, rt 70 min | OAc | OH | 79 |
| **78** | $MgCl_2$·THF (7.0 eq.) | ether rt 5h | Cl | OH | 99 |
| **79** | $MgBr_2$·$Et_2O$ (4.2 eq.) | ether rt 1h | Br | OH | 95 |
| **80** | $PhCH_2S^-Na^+$ (1.2 eq.) | DMF rt 4h | $SCH_2Ph$ | OH | 81 |
| **81** | $NMe_3$ (29 eq.) | THF -5 °C, 8h | $NMe_3^+$ | $O^-$ | 56 |
| **82** | $NaOCH_3$ (0.1 eq.) | THF/$CH_3OH$, rt 1h | OH | $OCH_3$ | 88 |

These results are analogous to those previously reported for the mono N-protected β-lactone **55**.[48,52] The reaction times are generally longer though, as is to be expected on steric grounds. While "hard" nucleophiles like methoxide attack the β-lactone with fission of the acyl oxygen-carbon bond, softer nucleophiles like sulphur, carboxylate, and halides cause alkyl oxygen-carbon cleaveage. Ammonia reacts by both pathways. The results show optically active compounds are produced, and although their

stereochemical purity has not been determined, analogy to reactions of the mono N-protected β-lactone **55** suggests that no epimerization occurs with the possible exception of the methoxide cleavage.[48]

The reaction of ammonia with β-lactone **64** in THF gives a 2:3 ratio of amine/amide products **75** and **76**, respectively. In the case of the monosubstituted β-lactone **55**, this ratio was 3:1 in THF and 1:3 in $CH_3CN$.[48] These results suggest that the energy difference between the two transition states leading to amine and amide products is not very great. Thus slight changes in solvents or structure can substantially alter the amine/amide product ratio. The attack of amines on β-propiolactone has been finely tuned by using amino trimethylsilyl, germanyl, stannyl, and arsine compounds.[135-137]

The products of these β-lactone ring openings are N-protected forms of important amino acids. For example, compound **75** is a convenient precursor not only for the preparation of α,β-diaminopropionic acid[138,139] but also for antibiotic and antitumor peptides containing it.[140-142] The β-O-acetyl **77**, β-chloro **78**[143,144] and β-bromo **79** compounds could in principle be converted to suicide substrates of bacterial enzymes.[145-147] Reaction of the β-lactone with benzylmercaptide affords a triprotected cysteine derivative **80** which could be useful in preparing cysteine containing peptides. The reaction with $MgBr_2$ is almost instantaneous whereas $MgCl_2$ reacts much slower. These results could have implications in deciding whether to use a bromo or chloromagnesium Grignard reagent in copper catalyzed ring openings.

Having shown halide, sulphur, oxygen and nitrogen nucleophiles attack the β-lactone **64** the possiblity of forming carbon-carbon bonds using organometallic reagents was investigated. Indeed higher order organocuprates (derived from 2 equivalents of organolithium and 1 equivalent of cuprous cyanide),[125] classical Gilman reagents (from 2 equivalents of organolithium and 1 equivalent of copper (I) bromide-

dimethyl sulfide complex),[148] and halomagnesium organocuprates (from 2 equivalents of Grignard reagent and one equivalent of cuprous bromide dimethyl sulfide complex) [149] opened the β-lactone **64**. The results are summarized in Table II.

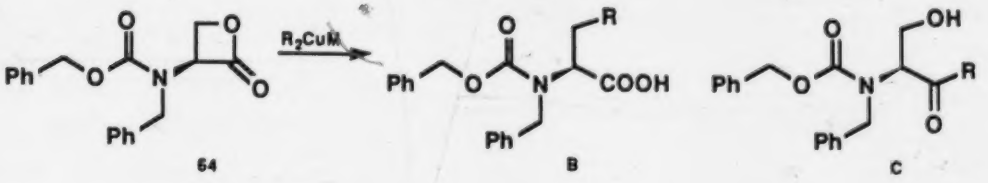
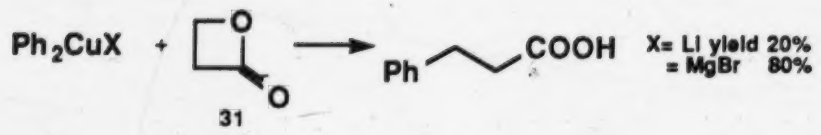



TABLE 2. Reaction of N-Benzyl-N-Benzyloxycarbonyl-α-Amino Serine β-Lactone (**64**) with Organocuprates.

| CuX | RM | Product B | Yield | Product C | Yield | ZNHBn **89** |
|---|---|---|---|---|---|---|
| $CuBr{\cdot}Me_2S$ | MeLi | **83** | 70 | | | |
| CuCN | MeLi | **83** | 82 | **84** | 5 | |
| $CuBr{\cdot}Me_2S$ | PhMgBr | **85** | 60 | | | |
| CuCN | PhLi | **85** | 25 | | | |
| CuCN | *n*-BuLi | **86** | 76 | **87** | 5 | |
| CuCN | *sec*-BuLi | **88** | 76 | | | 4 |
| $CuBr{\cdot}Me_2S$ | *t*-BuLi | **90** | 51 | | | 18 |
| CuCN | *t*-BuLi | **90** | 38 | | | 19 |

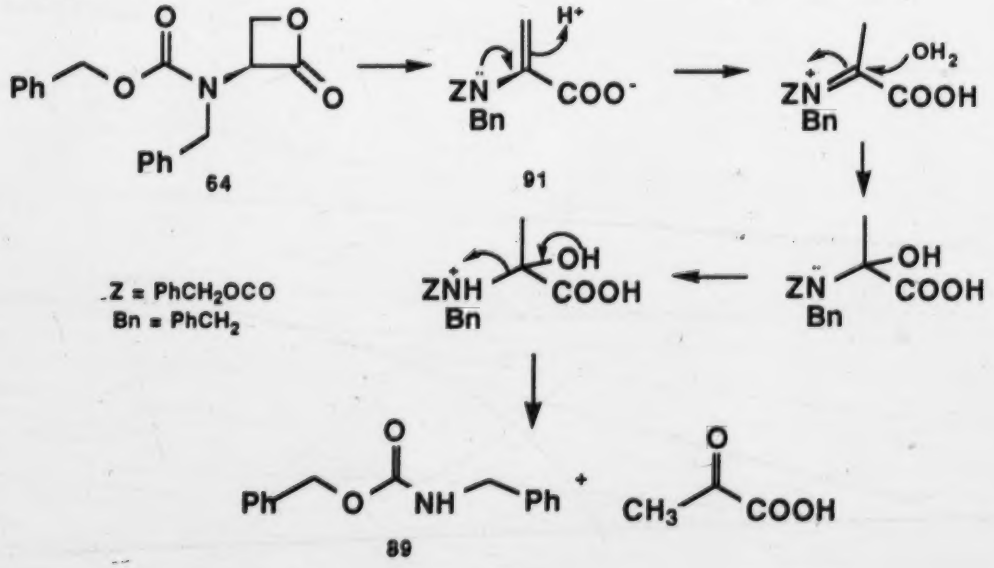
The Gilman reagent $Me_2CuLi{\cdot}Me_2S$[148] and Lipshutz reagents[125] $Me_2CuCNLi_2$ and *n*-$Bu_2CuCNLi_2$ give small amounts of the undesired attack at the the carbonyl. When the higher order cuprate, $Ph_2CuCNLi_2$, was replaced with $Ph_2CuMgBr$ the yield of N,N-diprotected phenylalanine **85** was improved from 25% to 60%. An analogous improvement was observed by Fujisawa and coworkers in the ring opening of β-propiolactone (**31**).[40]



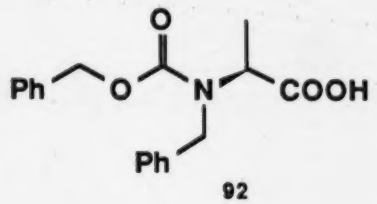

The $Ph_2CuCNLi_2$ has been shown to be quite effective in opening oxiranes, in displacements of primary halides, and in conjugate additions to α, β-unsaturated esters,[150] but does not give good yields with secondary halides, some α, β-unsaturated ketones,[125,150] or some acetates.[33]

Benzyl N-benzyl carbamate (**89**) was a side product in the *n*-$Bu_2CuCNLi_2$ reaction, and was formed to an even greater extent with both the higher order Lipshutz and the Gilman *t*-butyl reagent attack on β-lactone **64.** Compound **89** may result from base abstraction of the α-proton of the β-lactone with elimination to give the salt of N-benzyl-N-carbobenzyloxy dehydroalanine **91**; hydrolysis during the acidic workup would give pyruvic acid and benzyl N-benzyl carbamate (**89**) (Scheme 5).

Scheme 5

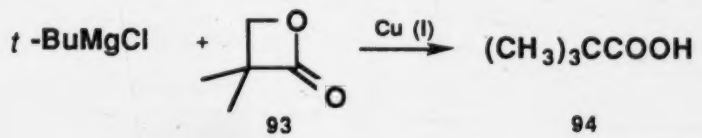


Another side product from reaction of **64** with *tert*-butyl organocuprates is the N,N-diprotected alanine **92** which results from hydride transfer to the β-methylene




**92**

carbon of **64**. Sato *et al*[39] noted that copper catalyzed ring opening of α, α-dimethyl-β-propiolactone (**93**) with *t*-BuMgCl produced pivalic acid (**94**) as a major product.



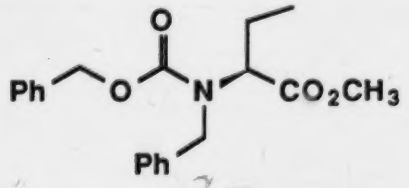

**93** **94**

The best yield O'Donnell and Falmagne observed for $(t\text{-Bu})_2Cu(CN)Li_2$ attack on iminium acetate **14** to form *tert*-leucine was 54%.[33]

Cuprous cyanide offers several advantages over the Cu(I) halides as the source of copper. This compound is not light sensitive or hygroscopic, and it is 14 to 19 times less expensive than the Cu(I) halides. In addition, the Cu(I) oxidation state is the more highly stable in CuCN[151] and the commercially available material does not need to be purified before use.

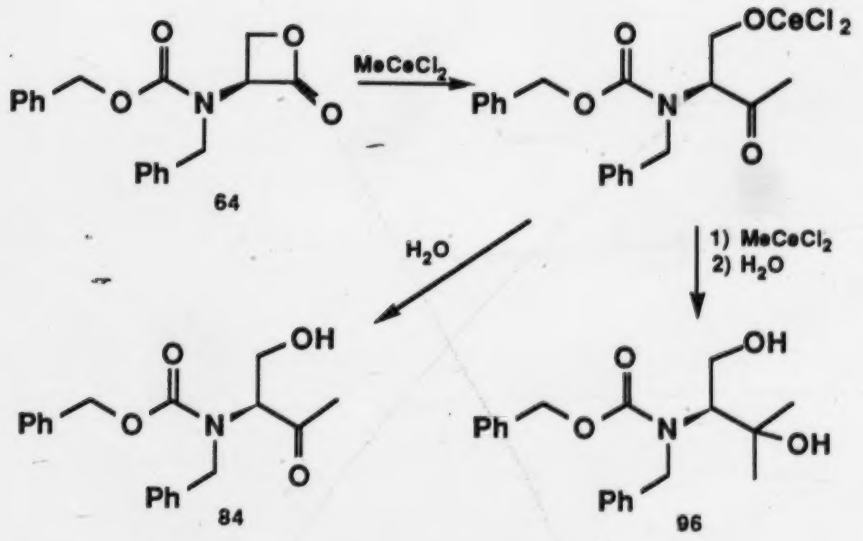
Purification of the products was complicated by their complexation with copper (II) ions resulting from air oxidation during workup of the reaction. Usual chromatographic procedures fail to remove this ion completely. Although, ion-exchange chromatography with Chelex resin removes the copper, it also appears to bind the products via a copper complex. Washing an ethyl acetate solution of the crude

products with a saturated solution of ethylenediaminetetraacetic acid (EDTA) removes the copper effectively. An alternative method could involve conversion of the crude acids to their benzyl esters with phenyldiazomethane and purification by flash chromatography. The purified products could then be hydrogenated to remove the carbobenzyloxy and two benzyl groups in a single step to afford the free amino acids. If the methyl esters were required phenyldiazomethane could be replaced by diazomethane. Addition of an etheral solution of diazomethane to a crude sample of acid **83** gave, after flash chromatography, an analyically pure sample of N,N diprotected α-aminobutyric acid methyl ester **95**.

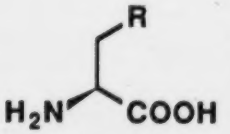



**95**

In an attempt to eliminate side products obtained in the cuprate reactions and to examine the possiblility of using more readily available "acidic" monoprotected β-lactones, organocerium reagents were investigated. Imamoto and coworkers had shown that organocerium (III) reagents generated by the reaction of one equivalent of organolithium or Grignard with cerium (III) chloride are extremely useful in providing addition products of easily enolizable ketones.[152,153] These results are ascribed to the reduced basicity of these organoceriums and the strong oxaphilicity of trivalent cerium.[154] The N, N-diprotected-β-lactone **64** did not react at the methylene but rather at the carbonyl carbon. Reaction of **64** with one equivalent of $MeCeCl_2$ afforded the corresponding keto alcohol **84** as well as the diol **96** resulting from a second addition to this product. The remainder of the isolated material was the starting β-lactone.

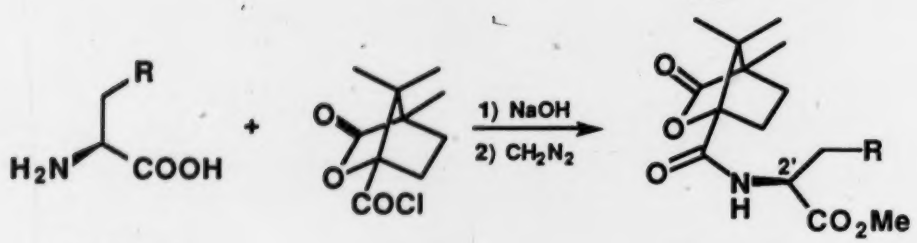



Hydrogenation (1atm) of the N, N-diprotected amino acids **83**, **85** and **90** in an acidic solvent system (ether, isopropyl alcohol, aqueous HCl) with 10% palladium on charcoal catalysis cleanly removed both protecting groups. The residue was purified by ion exchange chromatography to afford the free L-amino acids **97-99** in high yield (79-99%).




**97** R = $CH_3$
**98** R = $C_6H_5$
**99** R = t-Bu

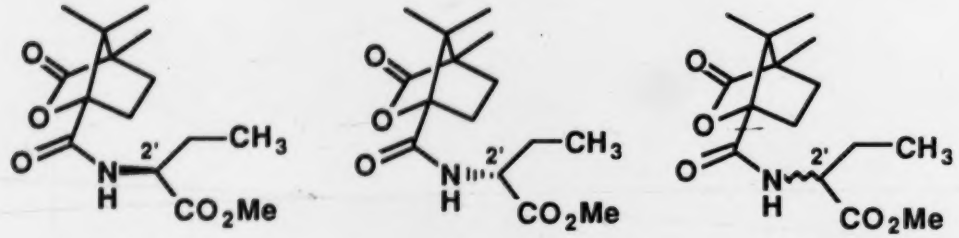
The stereochemical purity of two of these amino acids was determined by conversion to the corresponding camphanamide methyl esters and gas chromatography. This was accomplished by treatment of the amino acids **97** and **98** with (1S,4R)-camphanoyl chloride by the procedure of Armarego *et al* [155] followed by reaction with diazomethane to give **100** and **101**, respectively.




97 R = $CH_3$
98 R = $C_6H_5$

100 R = $CH_3$
101 R = $C_6H_5$

Authentic 2'S isomer **102**, authentic 2'R isomer **103**, and an equal mixture of both these diastereomers **104** was prepared from commercially available 2S, 2R, and 2RS α-aminobutyric acid (**105**, **106**, and **107**, respectively). Although the 400 MHz $^1H$ NMR spectra




102 103 104

of **102-104** did not display major differences, determination of enantiomeric purity proved to be possible by gas chromatography. The individual isomers were injected separately and as mixtures of defined isomeric content. The column (Alltech 10m x 0.53 mm bonded FSOT polyphenylmethylsiloxane (RSL-300)) conditions were 150 °C for 1 min then increasing 5 °C/min to 240 °C at a nitrogen carrier gas pressure of 8.5 psi. Table 3 summarizes the results.

TABLE 3 Gas Chromatographic Retention Times of 2' Isomers of Camphanamide Methyl Esters of α-Aminobutryic Acid.

| Entry | Diasteromer(s) | Retention Time (min) | |
|---|---|---|---|
| 1 | **100** | 11.88 | 12.31 |
| 2 | **102** | | 12.24 |
| 3 | **103** | 11.84 | |
| 4 | **104** | 11.82 | 12.24 |
| 5 | **100** and **102** | 11.85 | 12.29 |
| 6 | **100** and **103** | 11.85 | 12.25 |

The material prepared by opening of β-lactone **64** with $Me_2CuCNLi_2$ was shown to have an enantiomeric excess of 82%.

Analysis of phenylalanine **98** obtained by hydrogenation of **85** proceeded in a similar fashion. A 2:1 mixture **108** of 2'R:2'S diastereomers of the camphanamide methyl esters of this amino acid was prepared from authentic D- and L-phenylalanine. The column (same as above) conditions were 170 °C for 1 min then increasing 5 °C /min to 240 °C and remaining there for 10 min. The nitrogen carrier gas pressure was 8.5 psi. The results are shown in Table 4.

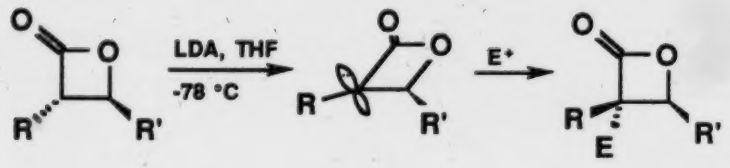
TABLE 4 Gas Chromatographic Retention Times of 2' Isomers of Camphanamide Methyl Esters of Phenylalanine.

| Entry | Diasteromer(s) | Retention Time | |
|---|---|---|---|
| 1 | **101** | 17.03 | 17.46 |
| 2 | **108** | 17.09 | 17.51 |
| 3 | **101** and **108** | 17.08 | 17.52 |

Integration of the peaks for the compound **101**, derived from Lipshutz reagent ring opening of β-lactone **64**, shows it to be 94% optically pure. The results demonstrate

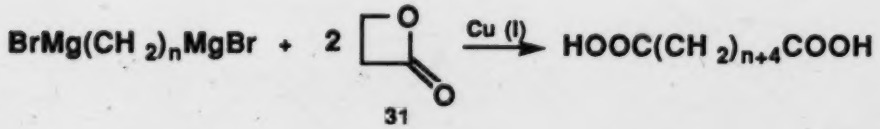
that a small amount of epimerization occurs during the ring opening reaction with highly basic organometallic reagents.

Mulzer has reported that β-lactones can be deprotonated with lithium diisopropyl amide (LDA) to give surprisingly stable anions.[122,156] Since the lone pair of the anion is orthogonal to the bond between the ring oxygen and the β-carbon, elimination is slow. Such anions can be alkylated in high yield with good diastereoselectivity if there are substituents on the β-carbon.



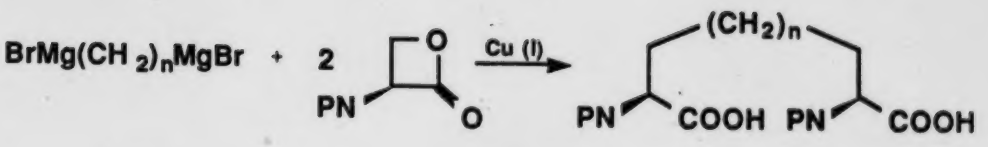

If this process could be repeated with α-amino-β-lactones then α-substituted α-amino acids would be generated. These non-proteinogenic amino acids often have important biological properties.[8,157,158]

Another report which may prove useful in amino acid synthesis via α-amino-β-lactones, is the observation that α,ω-di-Grignard reagents react with 2 equivalents of β-propiolactone (**31**) to give α,ω-dicarboxylic acids in modest yields with copper catalysis.[54]

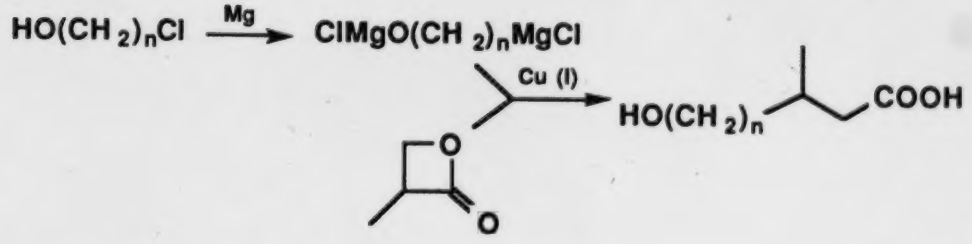



31

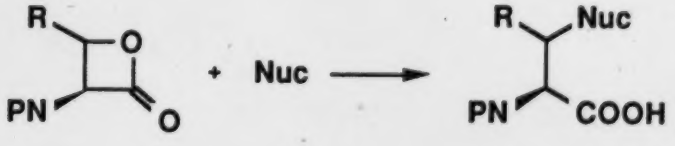
With α-amino-β-lactones, diaminodicarboxylic amino acids would be formed.




Though the reaction of β-lactones with alkoxide at the acyl carbon is well known,[50] ω-metaloxylated Grignard reagents react selectively with β-propiolactones in the presence of copper catalysis to give ω-hydroxycarboxylic acids.[55]



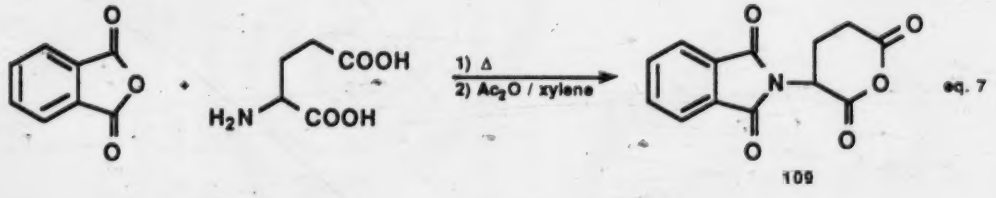

Extension of this methodology to higher homologs of serine such as threonine could prove to be a fruitful area of research as well.




Malonate addition to α-amino-β-lactones to prepare γ-substituted glutamic acids has been attempted in our laboratory.[114] As of yet reaction conditions to selectively open at the methylene carbon of N-benzyloxycarbonyl-β-lactone or N-*t*-butoxycarbonyl-β-lactone in synthetically useful yields have not been found. The reaction of lithium, sodium, and potassium dibenzyl and di-*tert*-butyl malonates produces primarily products from elimination to give dehydroalanine derivatives and

from attack at the carbonyl to give acyl adducts. Work is continuing in this area to provide the γ-substituted glutamic acids required for our work with thalidomide.

As a part of the study of the mechanism of action of thalidomide, several standards were required. We chose thalidomide (**36**), phthalimidino glutarimide (**37**) and N-phthalimidophthalimide (**42**). A common intermediate in the preparation of thalidomide (**36**) and phthalimidino glutarimide (**37**), phthaloyl glutamic anhydride (**109**), was prepared by heating a mixture of phthalic anhydride and L-glutamic acid to a melt and then cyclizing the resulting diacid to the anhydride with acetic anhydride[159] (equation 7).

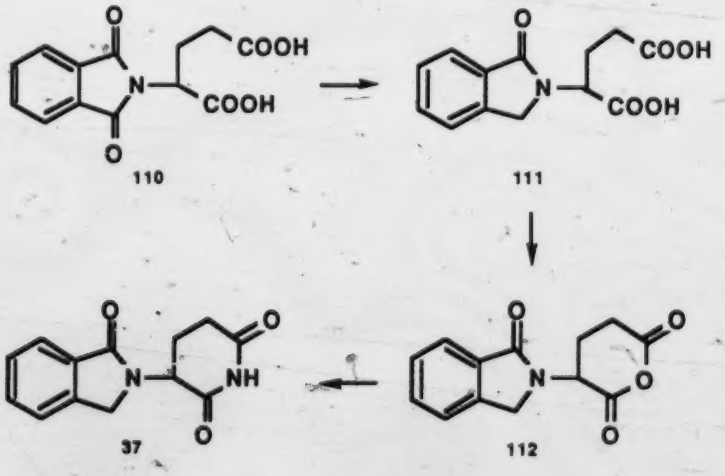



109

Heating the anhydride **109** with urea gave thalidomide (**36**).[97]

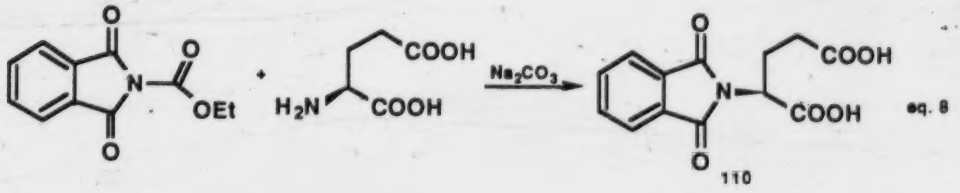
Reduction of the phthaloyl group to a phthalimidine was required for the preparation of **37** by the sequence shown in Scheme 5.

Scheme 5



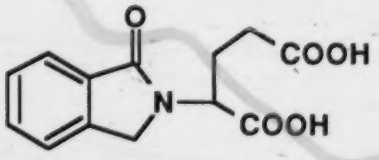

110 111 37 112

Phthaloyl glutamic acid **110** was prepared either by refluxing the anhydride **109** in water[160] or by condensation of N-carboethoxyphthalimide with L-glutamic acid under mild basic conditions[161] (equation 8).



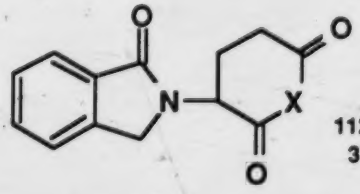

110 eq. 8

Clemmensen reduction of **110** with Zn dust in refluxing acetic acid afforded the phthalimidine **111** in 63% yield. Cyclization of this material to the anhydride **112**



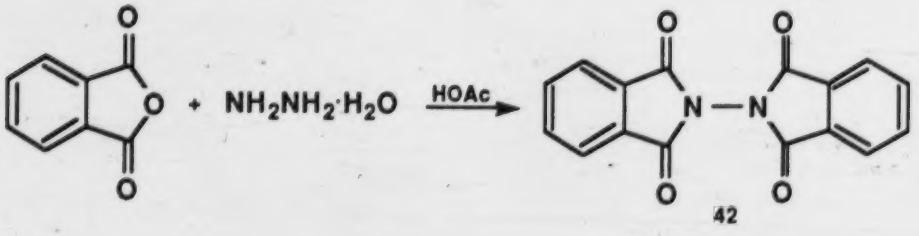

111

with acetic anhydride followed by imide formation with urea gave teratogen phthalimidinoglutarimide (**37**)[97] in 38% yield from **111** over these 2 steps.




112 X = O
37 X = NH

Another known teratogen N-phthalimidophthalimide **42**[93] was assembled by adding an excess of hydrazine hydrate to phthalic anhydride in refluxing acetic acid[162] as shown in equation 9.

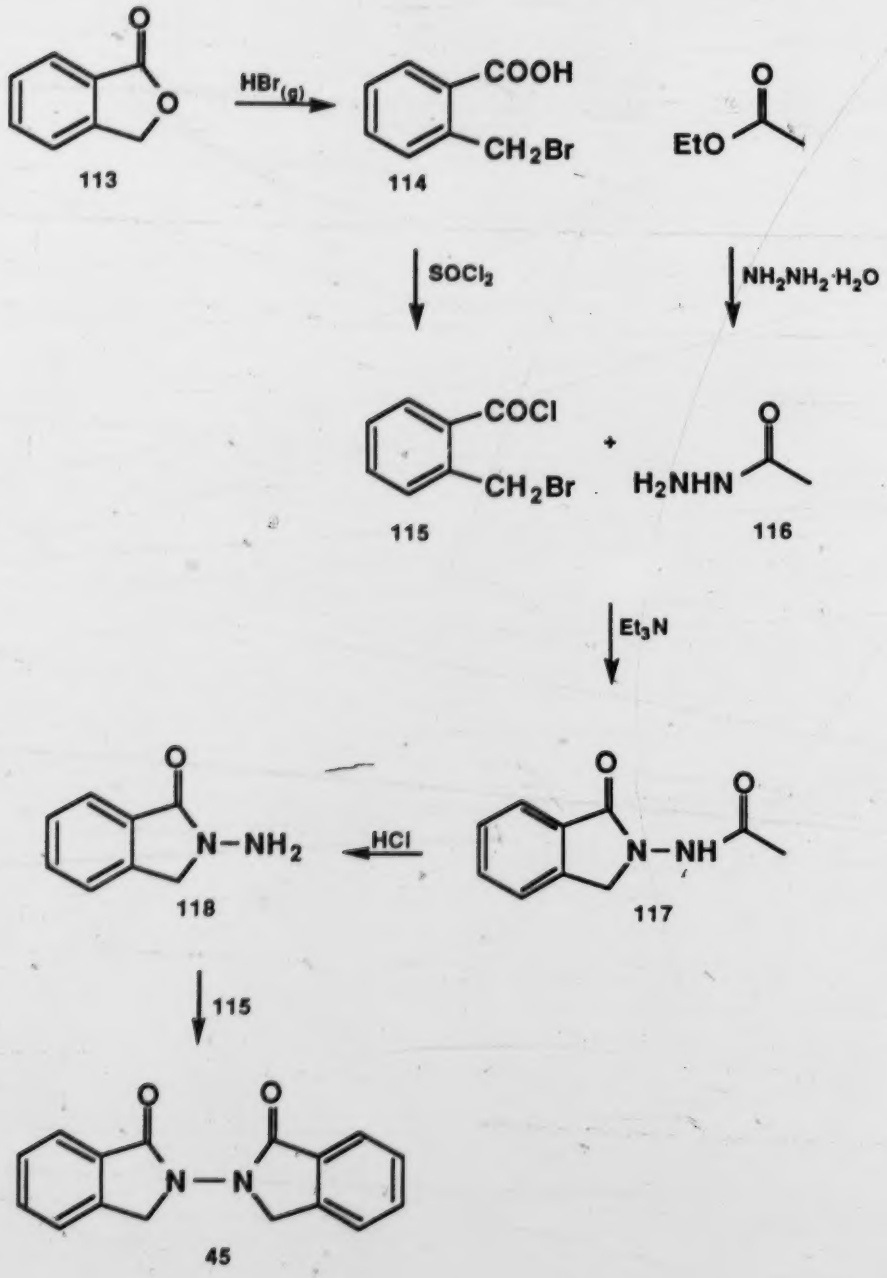



eq. 9

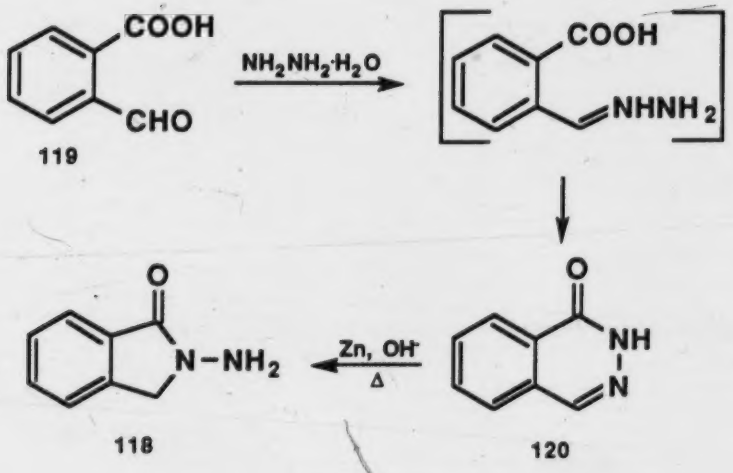
**42**

N-Phthalimidinophthalimidine **45** had been prepared previously by Bellasio and Testa[112] (Scheme 6) but has never been tested as a teratogen. This procedure was repeated with slight modification. The lactone ring of phthalide (**113**) was opened with hydrobromic acid.[163] Treatment of the resulting acid **114** with thionyl chloride afforded *o*-(bromomethyl)-benzoyl chloride (**115**) in 56% over these two steps. The extremely hygroscopic acetohydrazide (**116**) (prepared by condensation of hydazine hydrate with ethyl acetate[164]) was allowed to react with acid chloride **115** to form acetate **117**.[112] Hydrolysis of the acetyl protecting group with refluxing concentrated hydrochloric acid afforded the hydrochloride salt of **118**.[112] A more efficient route to **118** used *o*-formylbenzoic acid (**119**)[165] or commercially available phthalazone (**120**) as a starting material[166] (Scheme 7). Hydrazine hydrate was added dropwise to *o*-formylbenzoic acid (**119**). The reaction only took a few minutes, passing from a yellow solution due to the formation of the hydrazone, to an analytically pure white solid, phthalazone (**120**).[166] Clemmensen reduction of **120** in acidic media afforded phthalimidine whereas the hydrazide **118** was produced in 71% yield under basic

Scheme 6

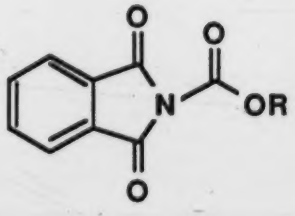

HBr(g)
COOH
CH2Br
EtO
113
114
SOCl2
NH2NH2·H2O
COCl
CH2Br
H2NHN
115
116
Et3N
N–NH2
HCl
N–NH
118
117
115
N–N
45

Scheme 7.



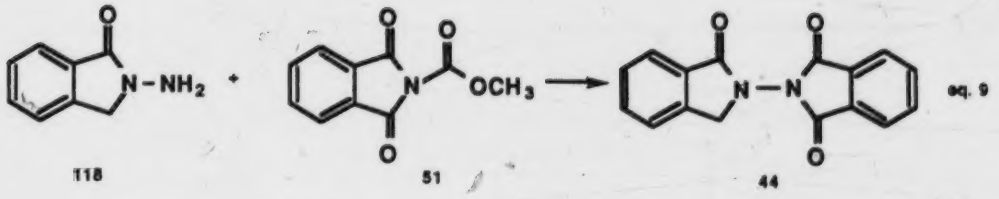

conditions.[166] Coupling of **118** with acid chloride **115** generated the desired N-phthalimidinophthalimidine (**45**) in 37% yield[112] (Scheme 6).

The analog **44** could be formed by coupling the hydrazide **118** with a suitable phthaloylating reagent. Work with N-phthaloyl-L-serine **46** suggested use of a reagent of the type shown below.




**51** R = $CH_3$
**121** R = $CH_2Ph$
**122** R = $C_6H_5$

Compound **51** was chosen because this allows removal of the methyl carbamate which is formed during the reaction by sublimation. Indeed coupling of **118** with **51** did produce **44** (61% yield) (equation 9).




118 51 44 eq. 9

These materials provide the standards for future teratological studies. They should prove especially useful if the nucleophilic opening of β-lactones can be eventually adapted to produce substituted glutamic acid derivatives which can be incorporated into thalidomide to further probe the mysteries of this biologically curious molecule.

## EXPERIMENTAL

All melting points were determined on a Thomas Hoover capillary melting point apparatus using open capillary tubes, and are uncorrected. Infrared spectra (IR) were recorded on a Nicolet 7199 FT-IR and mass spectra (MS) were obtained on A.E.I. MS-50 for electron impact (high resolution), MS-12 for chemical ionization (CI), or MS-9 for fast atom bombardment (FAB) instruments at an ionizing voltage of 70eV. Proton nuclear magnetic resonance ($^1$H NMR) spectra were recorded on Bruker WP-80 (80 MHz), Varian HA-100 (100 MHz), Bruker WH-200 (200 MHz), Bruker AM-300 (300 MHz), Bruker WM-360 (360 MHz) and Bruker WH-400 (400 MHz) instruments in the specified deuterated solvent with tetramethylsilane (TMS) or deuterated sodium 3-(trimethylsilyl)-1-propanesulfonate (TSP) as internal standards. Abbreviations used for NMR spectra are: s=singlet, d=doublet, t=triplet, q=quartet, m=multiplet, and br=broad. Carbon nuclear magnetic resonance ($^{13}$C NMR) spectra were recorded on Bruker HFX-90 (22.6 MHz), Bruker AM-300 (75.5 MHz) or Bruker WH-400 (100.6 MHz) instruments with TMS as internal, or 1,4-dioxane ($\delta$ 67.4 in $D_2O$) as external, reference standards. Optical rotation measurements were determined on a Perkin-Elmer 241 polarimeter in a 1 ml, 1 dm cell at ambient temperature (25 ± 2 °C).

Silica gel for column chromatography was Merck type 60, 70-230 mesh. Flash chromatography was carried out by the method of Still *et al*[167] using Merck type 60 silica gel, 230-400 mesh. Commercial thin layer chromatography (TLC) plates were silica gel, Merck 60F-254, or for reverse phase Whatman KC8F and Merck RP-8 F254s. TLC plates were visualized by quenching of fluorescence (254 nm) and/or staining with iodine, bromocresol green spray (acids), ninhydrin (amino acids), or sulfuric acid spray with charring. Reverse phase medium pressure liquid chromatography (MPLC) was performed using two Lichroprep RP-8 (40-63 μm)

columns in series. The first was a size A (240 mm x 10 mm) Lobar prepacked column and the second was a size B (310 mm x 25 mm). The acetonitrile eluant was distilled before use. Ion exchange chromatography employed Bio-Rad AG® 50W-X8 cation exchange resin (50-100 mesh).

Gas chromatographic analyses were performed on a Hewlett-Packard 5890A gas chromatograph equipped with a flame ionization detector. An Alltech 10 m x 0.53 mm bonded FSOT polyphenylmethylsiloxane (RSL-300) column was used with nitrogen as the carrier gas. All samples were injected as solutions in ethyl ether.

Solvents were dried, by distillation, under a static argon atmosphere from suitable desiccants. Dry benzene, ethyl ether, tetrahydrofuran (THF), and dioxane were distilled before use from sodium/benzophenone. Methanol and ethanol were distilled from magnesium, dichloromethane from calcium hydride, and acetonitrile from phosphorous pentoxide. Chloroform was washed with water, dried with sodium sulfate and then distilled from phosphorous pentoxide. Dimethylformamide (DMF) was stirred with barium oxide overnight and distilled under reduced pressure. Triethylamine was stored over 4Å molecular sieves. Triphenylphosphine was dried *in vacuo* for at least 16 h prior to use. Magnesium turnings and powder were dried at 120 °C overnight. Cuprous cyanide was dried overnight *in vacuo* at 56 °C over phosphorous pentoxide. Copper (I) bromide-dimethylsulfide complex was prepared by the method of Townsend and Theis.[124] Cuprous iodide (Fisher) was purified by literature methods.[123] Commercial (Aldrich) organolithium reagents were titrated before use by the menthol-phenanthroline method.[168] All other commercially-available reagents were used without purification unless otherwise noted.

Experiments requiring an inert dry atmosphere were performed under a slight positive pressure of argon with apparatus dried at 120 °C for at least three hours. Solvents and liquid reagents were transferred with syringes or double headed cannulas.

After extraction all organic layers were dried over anhydrous sodium sulfate except where otherwise indicated. Solvents were removed *in vacuo* on a Büchi rotary evaporator. Hydrogenation catalysts were removed by filtration through a small bed of Celite.

**N-Carbomethoxyphthalimide (51)**

The procedure of Nefkens *et al*[113] was followed. Methyl chloroformate (13.0 mL, 15.9 g, 0.168 mol) was added dropwise to a cooled (0 °C) suspension of potassium phthalimide (30.4 g, 0.164 mol) in DMF (100 mL) under an argon atmosphere. The mixture was stirred for 1 h at room temperature and poured onto 1 L of ice. The resulting solid was collected and recrystallized from 98% ethanol to give 19.2 g (57%) of **51**, m.p. 183-184.5 °C (lit.[113] m.p. 183 °C); IR ($CHCl_3$) 2960, 1810, 1776, 1756, 1718, 1310, 1259, 715 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 300 MHz) δ 7.97 (m, 2H, *Ar*), 7.83 (m, 2H, *Ar*), 4.03 (s, 3H, $CH_3$); exact mass 205.0372 (205.0375 calc. for $C_{10}H_7NO_4$); Anal. Calc. for $C_{10}H_7NO_4$: C, 58.54; H, 3.44; N, 6.83. Found: C, 58.38; H, 3.31; N, 6.81.

**N-Phthaloyl-L-serine (46)**

The procedure of Nefkens *et al*[113] was used with slight modification. Compound **51** (5.00 g, 24.4 mmol) was added to a solution of L-serine (**50**) (2.61 g, 24.8 mmol) in 1 M $Na_2CO_3$ (25 mL). This mixture was stirred for 1 h, filtered, and the filtrate was acidified to pH 1.5 with HCl. The resulting oil solidified at 4 °C overnight. These crystals were filtered and washed with cold water to give 1.65 g (29%) of **46**, m.p. 134-136 °C, $[\alpha]_{546}^{24}$ = -51.9°, c = 0.99, $CH_3OH$ (lit.[169] m.p. 135-136 °C, $[\alpha]_{546}^{21}$ = -50, c = 1, $CH_3OH$); IR ($CHCl_3$) 3200, 1777, 1712, 1393, 720 $cm^{-1}$; $^1H$ NMR ($CD_3CN$, 300 MHz) δ 7.86 (s, 4H, *Ar*), 6.07 (br s, 2H, O*H*),

4.95 (t, J = 7 Hz, 1H, C*H*), 4.10 (d, J = 7 Hz, 2H, C$H_2$O); exact mass 235.0481 (235.0481 calc. for $C_{11}H_9NO_5$); Anal. Calc. for $C_{11}H_9NO_5$: C, 56.17; H, 3.86; N, 5.96. Found: C, 55.97, H, 3.86; N, 5.74.

**N-Phthalimidophthalimide (44)**

The procedure of Drew and Hatt [162] was used. A solution of hydrazine hydrate (2.0 mL, 42 mmol) in glacial acetic acid (20 mL) was added over 30 min to a solution of phthalic anhydride (24.0 g, 162 mmol) in refluxing glacial acetic acid (40 mL). Heating was continued for 40 min, the mixture was cooled and the precipitate was filtered. The solid was washed with acetic acid and water, and was then stirred for 2 h with 0.25 N aqueous ammonia. The solid was filtered, boiled with water for 30 min, filtered again, and washed with ethanol. The precipitate was dried to give 7.17 g (58%) of **44**, m.p. 308-309 °C (lit.[170] m.p. 308-310 °C); IR ($CHCl_3$) 1781, 1740, 1468, 1345, 1280, 708 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 360 MHz) δ 7.99 (m, 4H, *Ar*), 7.86 (m, 4H, *Ar*); exact mass 292.0483 (292.0484 calc. for $C_{16}H_8N_2O_4$); Anal. Calc. for $C_{16}H_8N_2O_4$: C, 65.76; H, 2.76; N, 9.59. Found: C, 65.69; H, 2.85; N, 9.60.

**Preparation of L-serine Ethyl Ester Hydrochloride (52)**

L-Serine (**50**) (30.4 g, 0.290 mol) was suspended in anhydrous ethanol (1L) and HCl gas was added over 30 min at 0 °C. The mixture was allowed to warm overnight to 20 °C. The solution was concentrated *in vacuo*, redissolved in ethanol, and concentrated *in vacuo*. The residue was recrystallized from ethanol and ether to give 31.7 g (65%) of **52**, m.p. 129-131 °C, $[\alpha]_D^{25}$ = -4.2, c = 2.0, $H_2O$ (lit.[171] m.p. 130-132 °C, $[\alpha]_D^{20}$ = -4.4, c = 2, $H_2O$); IR (KBr) 3400, 3000, 1970, 1741, 1595, 1502, 1274, 1248, 1090, 1024 $cm^{-1}$; $^1H$ NMR ($D_2O$, 80 MHz) δ 4.37 (m, 3H, C*H*, C$H_2$Me), 4.20 (m, 2H, C$H_2$OH), 1.31 (t, J = 7 Hz, 3H, C$H_3$); MS (FAB) 134

($MH^+$); Anal. Calc. for $C_5H_{12}NO_3Cl$: C, 35.41; H, 7.13; N, 8.26. Found: C, 35.27; H, 7.08; N, 8.26.

**N-Trityl-L-serine Ethyl Ester (53)**

The procedure of Zervas and Theodoropoulos[117] was followed. Triethylamine (3.0 mL, 22 mmol) was added slowly to a cooled (0 °C) suspension of L-serine ethyl ester hydrochloride **52** (1.70 g, 10.0 mmol) in dry $CHCl_3$ (15 mL). Trityl chloride (2.79 g, 10.0 mmol) was added in portions, the mixture was stirred for 6 h, washed with water (3 x 8 mL), dried, and concentrated *in vacuo* to give 3.46 g (92%) of **53** as a foam which was used directly in subsequent reactions. A small sample slowly solidified on standing at 4 °C, m.p. 71-74 °C (lit.[172] m.p. 111-112 °C for racemic mixture); $[\alpha]_D^{25}$ = +6.0°, c = 1.0, $CHCl_3$; IR ($CHCl_3$) 3460, 3350, 3060, 2980, 1728, 1590, 1186, 1031, 747, 707 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.7-7.1 (m, 15H, *Ph*), 3.70 (m, 5H, C*H*, $CH_2OH$, $CH_2CH_3$), 2.98 (br s, 2H, O*H*, N*H*), 0.99 (t, J = 7 Hz, 3H, $CH_3$); exact mass 375.1824 (375.1835 calc. for $C_{24}H_{25}NO_3$); Anal. Calc. for $C_{24}H_{25}NO_3$: C, 76.77; H, 6.71; N, 3.73. Found: C, 76.87; H, 6.69; N, 3.67.

**N-Trityl-L-serine (24)**

The procedure of Guttmann[118] was followed with slight modification. Ester **53** (2.42 g, 6.45 mmol) was heated to reflux for 2 min in an ethanolic solution of 3.5 N KOH (2.0 mL, 7.0 mmol). The mixture was cooled to 4 °C, diluted with water (10 mL), and brought to pH 4 with 1 N $H_3PO_4$. The residue was filtered and washed with cold water ( 3 x 10 mL) to afford 1.31 g (58%) of **24**, m.p. 160 °C (dec.) (lit.[118] m.p. 160 °C (dec.) ); IR ($CH_2Cl_2$) 3320, 3055, 1596, 1493, 1030, 747, 705 $cm^{-1}$; $^1H$ NMR ($Me_2$ SO-$d_6$, 80 MHz) δ 7.60-7.15 (m, 15H, *Ph*), 4.20 (br s, 3H, COO*H*, O*H*, N*H*),

2.90 (m, 3H, C*H*, $CH_2$); exact mass 270.1129, 243.1174 (270.1131 calc. for $C_{15}H_{14}NO_3$ ($M^+$-Ph), 243.1174 calc. for $C_{19}H_{15}$ ($Ph_3C^+$))

**N-Trityl-L-serine β-Lactone (25)**

Compound **25** was prepared following the procedure of Shanzer and Libman.[65] N,N-Dimethylaminopyridine (0.231 g, 1.89 mmol) was added to a suspension of N-trityl-L-serine, (**24**) (3.12 g, 8.98 mmol) in dry $CH_2Cl_2$ (250 mL) at 0 °C. Diisopropyl carbodiimide (1.50 mL,, 9.60 mmol) was added dropwise and the mixture was stirred for 30 min at 0 °C and then 3 days at room temperature. The mixture was filtered. The solution was concentrated *in vacuo* and the residue was purified by flash chromatography (toluene) to give 0.290 g (10%) of **25**, m.p. 190-192 °C, $[\alpha]_D^{25}$ = -67°, c = 0.54, $CHCl_3$ (lit.[65] m.p. 193-194 °C, $[\alpha]_D^{28}$ = -62°, c = 0.5, $CHCl_3$); IR ($CHCl_3$) 3320, 3060, 1814, 1595, 1490, 1448, 1118, 873, 712, 704 $cm^{-1}$; $^1H$ NMR ($CD_2Cl_2$, 200 MHz) δ 7.50 (m, 6H, *Ph*), 7.29 (m, 9H, *Ph*), 4.60 (ddd, J = 4.7, 5.5, 11.5 Hz, 1H, C*H*) 3.56 (t, J = 5.5 Hz, 1H, $CH_2$), 3.14 (dd, J = 4.7, 5.5 Hz, 1H, $CH_2$) 2.71 (br d, J = 11.5 Hz, 1H, N*H*); MS (CI) 330 ($MH^+$).

**N-Benzyloxycarbonyl-L-serine β-Lactone (55)**

The procedure of Arnold *et al*[48] was used. Dimethyl azodicarboxylate[118] (4.40 mL, 40.1 mmol) was added dropwise over 15 min to a stirred solution of dry triphenylphosphine (10.5 g, 40.0 mmol) in anhydrous THF (200 mL) at -22 °C. After 20 min, a solution of N-carbobenzyloxycarbonyl-L-serine (**58**) (9.57 g, 40.0 mmol) in THF (230 mL) was added over 30 min. The mixture was stirred for 25 min at -22 °C and then 1 h at room temperature. Solvent was removed *in vacuo* and the residue was purified by chromatography on silica gel (80% hexanes/ethyl acetate; 55% hexanes/ethyl acetate) to give 3.11 g (35%) of β-lactone **55**, m.p. 131-132 °C, $[\alpha]_D^{26}$

= -26.9°, c = 1, $CH_3CN$ (lit.[48] m.p. 133-134 °C, $[\alpha]_D^{22}$ = -26.6°, c = 1, $CH_3CN$); IR ($CHCl_3$) 3360, 1845, 1829, 1686, 1531, 1268, 1104, 1016, 884, 753, 700 $cm^{-1}$; $^1H$ NMR ($CD_2Cl_2$, 80 MHz) δ 7.36 (s, 5H, *Ph*), 5.57 (br s, 1H, N*H*), 5.13 (s, 2H, OC$H_2$Ph), 5.01 (m, 1H, C*H*), 4.42 (d, J = 6Hz, 2H, C$H_2$); exact mass 221.0691 (221.0688 calc. for $C_{11}H_{11}NO_4$); Anal. Calc. for $C_{11}H_{11}NO_4$: C, 59.73; H, 5.01; N, 6.33. Found: C, 59.61; H, 5.03; N, 6.25.

**Dimethyl 1,2-Hydrazinedicarboxylate (57)**

The procedure of Kauer[121] was used. Methyl chloroformate (378 g, 4.00 mol) was added dropwise to hydrazine hydrate (100 g, 2.00 mol) in 95% ethanol (90 mL) at 0 °C. The addition was done in two equal portions, with the last half added concurrently with a solution of sodium carbonate (212 g, 2.00 mol) in $H_2O$ (800 mL). The reaction mixture was stirred for 3.5 h at room temperature. The resulting precipitate was collected and washed with cold water. A second crop of crystals was obtained by concentrating, and cooling the filtrate. The combined product was dried at 60 °C *in vacuo* and recrystallized from acetone/hexanes to give 190 g (64%) of **57**, m.p. 129-131 °C (lit.[121] m.p. 131-132 °C); IR ($CHCl_3$) 3306, 2960, 1720, 1525, 1237, 1068 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.83 (br s, 2H, N*H*), 3.71 (s, 6H, OC$H_3$); exact mass 148.0483 (148.0483 calc. for $C_4H_8N_2O_4$); Anal. Calc. for $C_4H_8N_2O_4$: C, 32.44; H, 5.44; N, 18.91. Found: C, 32.22; H, 5.39; N, 19.02.

**N-Benzyloxycarbonyl-DL-α-aminobutyric Acid(59)**

The procedure of Waley[173] was followed. Benzyl chloroformate (3.2 mL, 22 mmol) was added over 10 min to a cooled (0 °C) solution of DL-α-aminobutyric acid (2.07 g, 20.1 mmol) in 2 N NaOH (30 mL, 60 mmol). The reaction mixture was stirred for 3 h, washed with ether (2 x 35 mL), acidified to pH 2 with 1 N HCl, and

extracted with ether (3 x 40 mL). The dried ether phase was concentrated *in vacuo* and the residue was recrystallized from ether and petroleum ether (30-60) to afford 1.55 g (29%) of the N-benzyloxycarbonyl-DL-α-aminobutyric acid (**59**), m.p. 72-73.5 °C (lit.[174] m.p. 78-79 °C); IR ($CHCl_3$) 3320, 3100, 3030, 2970, 1718, 1529, 1230, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 10.47 (s, 1H, COO*H*), 7.35 (s, 5H, *Ph*), 6.35 (br s, 0.2H, N*H*), 5.45 (br d, J = 8 Hz, 0.8H, N*H*) 5.14 (s, 2H, $CH_2O$), 4.38 (m, 1H, C*H*), 1.85 (m, 2H, $CH_2$), 0.96 (t, J = 7 Hz, 3H, $CH_3$); exact mass 237.1001 (237.1001 calc. for $C_{12}H_{15}NO_4$); Anal. Calc. for $C_{12}H_{15}NO_4$: C, 60.75; H, 6.37; N, 5.90. Found: C, 60.91; H, 6.19; N, 5.99.

**N-Benzyloxycarbonyl-L-leucine (60)**

Benzyl chloroformate (3.5 mL, 25 mmol) was added to a cooled (0 °C) solution of L-leucine (2.62 g, 20.0 mmol) in 2 N NaOH (30 mL, 60 mmol). This mixture was stirred for 20 min at 0 °C, warmed to room temperature for 1.5 h, and washed with ether (2 x 15 mL). The aqueous layer was acidified with 5 N HCl and extracted with ether (200 mL). The ether phases were dried and concentrated *in vacuo* to give 2.91 g (55%) of N-benzyloxycarbonyl-L-leucine (**60**) as a colorless oil; IR ($CHCl_3$) 3315, 3100, 3030, 2960, 1719, 1531, 1266, 1230, 1050, 697 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 10.32 (s, 1H, COO*H*) 7.33 (s, 5H, *Ph*), 6.30 (br s, 0.25H, N*H*), 5.13 (br s, 2.75H, N*H*, $CH_2O$), 4.40 (m, 1H, C*H*COO), 1.68 (m, 3H, C*H*$CH_2$), 0.94 (d, J = 5 Hz, 6H, $CH_3$); exact mass 265.1317 (265.1314 calc. for $C_{14}H_{19}NO_4$); Anal. Calc. for $C_{14}H_{19}NO_4$: C, 63.38; H, 7.22; N, 5.28. Found: C, 63.42; H, 7.17; N, 5.16.

**N-Benzyloxycarbonyl-DL-phenylalanine (61)**

DL-Phenylalanine (3.30 g, 20.0 mmol) was treated with 2 N sodium hydroxide (30 mL, 60 mmol) and benzylchloroformate (3.5 mL, 4.2 g, 25 mmol), as described

above for the preparation of **60**, to give 3.52 g (59%) of **61**, m.p. 99-101 °C (lit.[175] m.p. 101 °C); IR ($CHCl_3$) 3320, 3100, 3030, 1719, 1518, 1498, 1455, 1259, 1216, 1955, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 300 MHz) δ 9.95 (s, 1H, COO*H*), 7.30 (m, 8H, *Ph*) 7.14 (m, 2H, *Ph*), 6.24 (br d, J = 8.5 Hz, 0.2H, N*H*), 5.25 (d, J = 8.5 Hz, 0.8H, N*H*), 5.10 (s, 2H, $CH_2$0), 4.70 (dd, J = 5.5, 6.5 Hz, 0.8H, C*H*), 4.51 (m, 0.2H, C*H*), 3.19 (dd, J = 5.5, 14 Hz, 0.8H, C*H*HPh), 3.09 (dd, J = 6.5, 14 Hz, 0.8H, CH*H*Ph), 2.94 (m, 0.4H, C*HH*Ph); exact mass 299.1168 (299.1158 calc. for $C_{17}H_{17}NO_4$); Anal. Calc. for $C_{17}H_{17}NO_4$: C, 68.22; H, 5.72; N, 4.68. Found: C, 68.20; H, 5.63; N, 4.56.

**Benzyl Carbamate (62)**

Benzyl chloroformate (3.0 mL, 21 mmol) was added to cold (0 °C) aqueous ammonia (2.4 N, 25 mL, 60 mmol). After 20 min, the reaction mixture was extracted with ether. The ether was dried with $MgSO_4$ and concentrated *in vacuo* to leave a white solid which was washed with petroleum ether to give 0.64 g (20%) of **62**, m.p. 85-86 °C (lit.[171] m.p. 87-89 °C); IR ($CHCl_3$) 3409, 3334, 3273, 3195, 3030, 2950, 1688, 1609, 1446, 1404, 1073, 753, 697 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.32 (s, 5H, *Ph*), 5.07 (s, 2H, $CH_2$), 4.90 (br s, 2H, $NH_2$); exact mass 151.0634 (151.0634 calc. for $C_8H_9NO_2$); Anal. Calc. for $C_8H_9NO_2$: C, 63.56; H, 6.00; N, 9.27. Found: C, 63.61; H, 5.88; N, 9.04.

**N-Benzyl-L-serine (65) from L-Serine Methyl Ester Hydrochloride (68)**

The procedures of Hardegger *et al*[128] and Velluz *et al*[129] were adapted. Chloroform saturated with ammonia gas (100 mL) was added to a suspension of L-serine methyl ester hydrochloride (**68**) (18.8 g, 0.121 mol) in $CHCl_3$ (100 mL) below -12 °C. The resulting solution was filtered through a Celite pad, concentrated *in vacuo*,

and treated with distilled benzaldehyde (11.5 mL, 0.113 mol) and sodium sulfate (21.6 g, 0.152 mol). The reaction mixture was stirred 2 h, ether was added, and the mixture was filtered and concentrated *in vacuo* . The residue was dissolved in anhydrous methanol (50 mL) and added to 3.1 g of 5% palladium on charcoal which had been prehydrogenated for 1 h at 29 psi. The mixture was stirred under hydrogen (20 psi.) for 6 h,and the catalyst was removed by filtration through a Celite pad. Saponfication with refluxing 3.6 N ethanolic potassium hydroxide (75 mL) for 15 min followed by acidification of the cooled solution to pH 5 with 6 N HCl gave a precipitate. This was filtered and washed with chloroform to afford 10.5 g (49%) of N-benzyl-L-serine **65**, m.p. 220 °C, $[\alpha]_{578}^{25}$ = +5.0°, c = 1.0, 6 N HCl (lit.[176] m.p. 220-222 °C (dec.), lit.[128] $[\alpha]_{578}$ = +5.1, c = 1, 6 N HCl); IR (KBr) 3260, 3000, 2820, 1642, 1593, 1552, 1067, 730, 695 $cm^{-1}$; $^1H$ NMR ($D_2O$, 80 MHz) δ 7.40 (s, 5H, *Ph*), 4.21 (s, 2H, $CH_2N$), 3.88 (d, J = 4.5 Hz, 1H, *CH*), 3.11 (t, J = 4.5 Hz, 2H, $CH_2O$); exact mass 195.0888 (195.0896 calc. for $C_{10}H_{13}NO_3$); Anal. Calc. for $C_{10}H_{13}NO_3$: C, 61.53; H, 6.71; N, 7.17. Found: C, 61.49; H, 6.73; N, 7.29.

**N-Benzyl-L-serine (65) from L-Serine and N-Benzyl-D-serine (67) from D-Serine**

The procedure of Quitt *et al*[130] was used. Distilled benzaldehyde (10.1 mL, 99.4 mmol) was added to L-serine (10.5 g, 99.9 mmol) in 2 N NaOH (50 mL). The solution was stirred for 40 min, cooled to 4 °C, and treated with small portions of sodium borohydride (1.13 g, 29.9 mmol). Stirring was continued for 1 h at room temperature after the addition was complete, and the procedure was repeated with the same quantities of benzaldehyde and sodium borohydride. The reaction mixture was extracted with ether (115 mL) to remove impurities. The aqueous phase was chilled (4 °C), acidified to pH 6 with 1 N HCl, and filtered. The residue was recrystallized from

water to give 6.95 g (36%) of **65**, m.p. 220-222 °C (dec.), $[\alpha]_{578}^{25}$ = +5.0°, c = 0.94, 6 N HCl (lit.[176] m.p. 220-222 °C (dec.), lit.[128] $[\alpha]_{578}$ = +5.1°, c = 1, 6 N HCl). The spectral properties are identical to those for **65** above.

The corresponding D-isomer **67** was prepared analogously: $[\alpha]_{578}^{25}$ = -5.0°, c = 1.0, 6 N HCl.

**N-Benzyl-L-serine Methyl Ester Hydrochloride (71)**

Crude N-benzyl-L-serine methyl ester (**70**) (2.20 g, 10.5 mmol) (from the preparation of N-benzyl-L-serine (**65**) from L-serine methyl ester hydrochloride (**68**) above) in anhydrous ether (5 mL) was treated with HCl gas.[128] Recrystallization of the resulting precipitate from acetone and ether gave 1.82 g (71%) of the hydrochloride salt **71**, m.p. 139-141 °C, $[\alpha]_{578}^{25}$ = -10.5, c = 1.7, absolute ethanol (lit.[128] m.p. 139 °C; $[\alpha]_{578}$ = -10.8, c = 1.8, absolute ethanol); IR (KBr) 3338, 1746, 1550, 1079, 757, 696 $cm^{-1}$; $^1H$ NMR ($D_2O$, 80 MHz) δ 7.42 (s, 5H, *Ph*), 4.30 (s, 2H, $CH_2N$), 4.20-4.05 (m, 3H, *CH*, $CH_2O$), 3.78 (s, 3H, $OCH_3$); MS (FAB) 210 ($MH^+$); Anal. Calc. for $C_{11}H_{16}ClNO_3$: C, 53.77; H, 6.56; N, 5.70. Found: C, 53.57; H, 6.53; N, 5.97.

**N-Benzyl-L-serine (65) from its Methyl Ester (71)**

The procedure of Hardegger *et al* [128] was followed. N-Benzyl-L-serine methyl ester hydrochloride (**71**) (25.1 g, 0.102 mol) was heated to reflux for 1 h with 2.3 M hydrochloric acid (250 mL). The solution was concentrated *in vacuo* , diluted with water (25 mL) and adjusted to pH 5 with 2 N LiOH to give a precipitate. Filtration and recrystallization from water afforded 13.0 g (65%) of **65** with spectral properties identical to those for **65** above, m.p. 220-222 °C (dec.); $[\alpha]_{578}^{25}$ = +5.1, c = 1.01, 6 N HCl, (lit.[176] m.p. 220-222 °C (dec.), $[\alpha]_{578}$ = +5.1, c = 1, 6 N HCl); Anal. Calc. for $C_{10}H_{13}NO_3$: C, 61.53; H, 6.71; N, 7.17. Found: C, 61.72; H, 6.79; H, 7.31.

**N-Benzyl-N-benzyloxycarbonyl-L-serine (72) and the D-Isomer 73**

Benzyl chloroformate (0.45 mL, 3.0 mmol) was added dropwise to a solution of N-benzyl-L-serine (**65**) (0.203 g, 1.03 mmol) in 0.23 M potassium bicarbonate (13 mL). This solution was stirred for 2 h and washed with ether (2 x 20 mL). The aqueous phase was acidified to pH 2 with 1 M HCl and extracted with $CHCl_3$ (3 x 20 mL). The $CHCl_3$ extracts were dried and concentrated *in vacuo* to afford 0.159 g (47%) of **72** as a colorless oil; $[\alpha]_D^{25}$ = -24.4°, c = 1.3, $CHCl_3$; IR ($CHCl_3$) 3300 , 3030, 1700, 1473, 1454, 1425, 1245, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.29 (s, 10H, *Ph*), 5.78 (br s, 2H, COO*H*, O*H*), 5.18 (s, 2H, PhC$H_2$O), 4.60 (br s, 2H, C$H_2$N), 4.25 - 3.60 (m, 3H, C$H_2$OH, C*H*); exact mass 329.1264 (329.1263 calc. for $C_{18}H_{19}NO_5$); Anal. Calc. for $C_{18}H_{19}NO_5$: C, 65.64; H, 5.82; N, 4.25. Found: C, 65.42; H, 5.72; N, 4.25.

For the D-isomer **73** prepared analogously: $[\alpha]_D^{25}$ = +24.3°, c = 1.3, $CHCl_3$.

**N-Benzyl-N-benzyloxycarbonyl-L-serine β-Lactone (64) and the D-Isomer 66**

Dimethyl azodicarboxylate (1.70 mL, 15.5 mmol) was added dropwise to a cooled (-78 °C) solution of dry triphenylphosphine (4.07 g, 15.5 mmol) in dry THF (60 mL) and the resulting mixture was stirred for 20 min. A solution of β-hydroxy acid, **64** (4.96 g, 15.1 mmol) in dry THF (130 mL) was added over 30 min. The mixture was stirred for 40 min at -78 °C, allowed to warm to room temperature, and then stirred for an additional 2 h. The solvent was removed and the residue purified by flash chromatography on silica gel (80% hexanes/ethyl acetate; 65% hexanes/ethyl acetate) to afford 3.34 g (71%) of β-lactone **64** which was recrystallized from ether: m.p. 73-74 °C, $[\alpha]_D^{26}$ = -9.3°, c = 1, THF; IR ($CHCl_3$) 3030, 1833, 1702, 1247, 1107, 1014, 889, 699 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.33 (m, 10H, *Ph*), 5.23

(s, 2H, PhC$H_2$O), 4.94 (dd, J = 5, 6.5 Hz, 1H, C*H*), 4.58 (s, 2H, C$H_2$N), 4.23 (m, 2H, CHC$H_2$O); exact mass 311.1157 (311.1157 calc. for $C_{18}H_{17}NO_4$); Anal. Calc. for $C_{18}H_{17}NO_4$: C, 69.44, H, 5.50; N, 4.50. Found: C, 69.44; H, 5.49; N, 4.44.

The corresponding D-isomer **66** was prepared analogously: $[\alpha]_D^{25}$ = +9.6°, c = 1, THF. The other spectral properties are analogous to those reported for **64** above.

**Reaction of Ammonia with 64 to form (S) $N^{\alpha}$-Benzyl-$N^{\alpha}$-benzyloxycarbonyl-2,3-diaminopropionic Acid (75) and $N^{\alpha}$-Benzyl-$N^{\alpha}$-benzyloxycarbonyl-L-serinamide (76)**

Ammonia gas was bubbled through a solution of β-lactone **64** (0.103 g, 0.330 mmol) in dry THF (5 mL) at 0 °C for 2 h, followed by addition for 3 h at room temperature. The mixture was stirred for 6 h, the solvent was removed *in vacuo* , and the residue was purified by flash chromatography on silica gel (70% ethyl acetate/ hexanes; 99% ethyl acetate/formic acid) to afford 39.7 mg (37%) of amine **75** and 54.9 mg (51%) of amide **76** as colorless oils. For **75**: $[\alpha]_D^{25}$ = -50°, c = 0.23, $CHCl_3$; IR ($CH_2Cl_2$) 3415, 3350, 3030, 1701, 1536, 1242, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 360 MHz) δ 7.28 (m, 10H, *Ph*), 5.13 (s, 2H, PhC$H_2$O), 4.8-3.4 (m, 8H, PhC$H_2$N, C*H*C$H_2$N$H_3$); exact mass 328.1405, 310.1078 (328.1423 calc. for $C_{18}H_{20}N_2O_4$, 310.1079 calc. for $C_{18}H_{16}NO_4$).

For **76**: $[\alpha]_D^{25}$ = -53°, c = 0.44, $CHCl_3$; IR ($CH_2Cl_2$) 3410, 3340, 3200, 3030, 1679, 1604, 1495, 1454, 1418, 1365, 1251, 698 $cm^{-1}$; $^1H$ NMR (360 MHz, $CDCl_3$) δ 7.35 (m, 10H, *Ph*), 6.2-5.4 (br m, 2H, N$H_2$), 5.21 (s, 2H, OC$H_2$Ph), 4.9-3.0 (br m, 6H, C$H_2$N, C*H*C$H_2$O*H*); exact mass 328.1423, 284.1288 (328.1423 calc. for $C_{18}H_{20}N_2O_4$, 284.1286 calc. for $C_{17}H_{18}NO_3$); Anal. Calc. for $C_{18}H_{20}N_2O_4$: C, 65.84; H, 6.14; N, 8.53. Found: C, 65.72; H, 6.21; N, 8.68.

**O-Acetyl-N-benzyl-N-benzyloxycarbonyl-L-serine (77)**

The β-lactone **64** (0.109 g, 0.349 mmol) was added to a solution of potassium acetate (0.241 g, 2.45 mmol) in DMF (11 mL) and water (0.5 mL). The mixture was stirred for 70 min, diluted with water (3 mL), cooled to 0 °C and acidified to pH 2 with 1 N HCl. This solution was extracted with $CH_2Cl_2$ and the extracts were concentrated *in vacuo* . The residue was dissolved in 0.5 N $KHCO_3$ (5 mL) and extracted with ethyl acetate. The aqueous layer was acidified to pH 2 with 1 N HCl and extracted with ethyl acetate. This ethyl acetate extract was dried and concentrated *in vacuo* to give 0.103 g (79%) of **77** as a colorless oil; $[\alpha]_D^{26}$ = -41.8°, c = 1.1 in THF; IR ($CHCl_3$ cast) 3100, 1744, 1704, 1230, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 200 MHz) δ 10.39 (br s, 1H, COO*H*), 7.22 (m, 10H, *Ph*), 5.14 (s, 2H, PhC$H_2$O), 4.74-4.26 (m, 5H, C$H_2$N, C*H*, C$H_2$OAc), 1.82 (s, 3H, OOCC$H_3$); MS (CI) 389 $MNH_4^+$; Anal. Calc. for $C_{20}H_{21}NO_6$: C, 64.68; H, 5.70; N, 3.77. Found: C, 64.61; H, 5.70; N, 3.64.

**N-Benzyl-N-benzyloxycarbonyl-β-chloro-L-alanine (78)**

Magnesium chloride was prepared according to Ashby and Arnott.[177] Magnesium powder (1.98 g, 81.4 mmol) was added to a solution of $HgCl_2$ (6.86 g, 25.3 mmol) in dry THF (25 mL) at 0 °C under argon. The mixture was heated to reflux for 2 h in the dark, stirring was discontinued, and 3.0 mL (3.0 mmol) of the supernatant $MgCl_2$ solution was added dropwise over 5 min to β-lactone **64** (0.136 g, 0.436 mmol) in dry diethyl ether (20 mL). The mixture was stirred for 5 h, cooled (0 °C), and acidified with 0.15 N $H_3PO_4$ to pH 3. The diethyl ether phase was separated and the aqueous phase was extracted with diethyl ether (3 x 15 mL). The combined extracts were washed with brine (15 mL), dried, and concentrated *in vacuo* to give 0.152 g of pure **78** (quantitative yield), $[\alpha]_D^{25}$ = -78.5°, c = 1.06, $CHCl_3$; IR ($CHCl_3$) 3100, 3035, 1709, 1454, 1425, 1242, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 8.50

(br s, 1H, COO*H*), 7.27 (s, 10H, *Ph*), 5.15 (s, 2H, PhC$H_2$O), 4.80 (d, J = 16 Hz, 1H, C*H*HN), 4.45 (d, J = 16 Hz, 1H, CH*H*N), 4.3-3.8 (m, 3H, CH, C$H_2$Cl); MS(FAB) 350, 348 ($MH^+$); Anal. Calc. for $C_{18}H_{18}ClNO_4$: C, 62.16; H, 5.22; N, 4.03. Found: C, 61.82; H, 5.08; N, 3.99.

**N-Benzyl-N-benzyloxycarbonyl-β-bromo-L-alanine (79)**

This material was prepared according to the procedure described above for the chloride **78**. The magnesium bromide solution was made using $HgBr_2$ (4.54 g, 12.6 mmol) in dry diethyl ether (12.5 mL) and magnesium powder (1.04 g, 42.8 mmol). This magnesium bromide solution (2.0 mL, 2.0 mmol) was added to β-lactone **64** (0.149 g, 0.477 mmol) in diethyl ether (20 mL). The mixture was acidified to pH 3 after 1 h and 0.178 g (95%) of bromide **79** was isolated as described for chloride **78** above. $[\alpha]_D^{25}$ = -60°, c = 0.28, $CHCl_3$; IR ($CHCl_3$) 3060, 3030, 1708, 1236, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 10.72 (br s, 1H, COO*H*), 7.29 (s, 10H, *Ph*), 5.18 (s, 2H, PhC$H_2$O), 4.88 (d, J = 16 Hz, 1H, C*H*HN), 4.50 (d, J = 16 Hz, 1H, CH*H*N), 4.27 (m, 1H, C*H*), 3.87 (br d, J = 7 Hz, 2H, C$H_2$Br); MS (FAB) 394, 392 ($MH^+$); Anal. Calc. for $C_{18}H_{18}BrNO_4$: C, 55.11; H, 4.63; N, 3.57. Found: C, 55.61; H, 4.71; N, 3.44.

**N-Benzyl-S-benzyl-N-benzyloxycarbonyl-L-cysteine (80)**

Benzylmercaptan (1.32 g, 10.6 mmol) was added dropwise over 10 min to sodium hydride (0.232 g, 9.66 mmol) suspended in dry N,N-dimethylformamide (DMF) (10 mL). The solution was stirred for 1 h and a portion (0.68 mL, 0.66 mmol) was added over 7 min to β-lactone **64** (0.178 g, 0.572 mmol) in dry DMF (10 mL). The reaction mixture was acidified after 4 h with 0.05 N $H_3PO_4$ and extracted with ethyl acetate (3 x 25 mL). The aqueous layer was concentrated *in vacuo* and the

residue was extracted with ethyl acetate (3 x 30 mL). The combined ethyl acetate phases were washed with brine (15 mL), dried, and concentrated *in vacuo* . The residue was purified by flash chromatography (30% ethyl acetate/hexanes; 99% ethyl acetate/acetic acid) to afford 0.203 g (81%) of **80** as a colorless oil; $[\alpha]_D^{25}$ = -57°, c = 0.38, $CHCl_3$; IR ($CHCl_3$) 3060, 1706, 1235, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 9.88 (br s, 1H, COO*H*), 7.25 (s, 15H, *Ph*), 5.17 (s, 2H, PhC$H_2$O), 4.8-4.1 (m, 3H, C$H_2$N, C*H*), 3.48 (br s, 2H, SC$H_2$Ph), 3.01 (m, 2H, C$H_2$SBn); exact mass 435.1495 (435.1504 calc. for $C_{25}H_{25}NO_4S$); Anal. Calc. for $C_{25}H_{25}NO_4S$: C, 68.94; H, 5.79; N, 3.22. Found: C, 68.64; H, 5.73; N, 3.30.

**N$^\alpha$-Benzyl-N$^\alpha$–benzyloxycarbonyl-β-(trimethylammonio)-L-alanine, Inner Salt (81)**

Trimethylamine (1.5 mL, 16 mmol) was added to a solution of β-lactone **64** (169 mg, 0.544 mmol) in THF (10 mL) at -78 °C. The solution was sealed, warmed to 0 °C, and stirred for 8 h. Removal of the solvent *in vacuo* provided a white powder which was recrystallized from methanol and ether (without heating) to give 112 mg (56%) of **81**, m.p. 116 °C (dec.); $[\alpha]_D$ = -17°, c = 0.52, $CH_3OH$; IR (MeOH) 1691, 1629, 1250, 1226, 1110, 698 $cm^{-1}$; $^1H$ NMR ($CD_3OD$, 360 MHz) δ 7.31 (m, 10H, *Ph*), 5.17 (s, 2H, C$H_2$O), 4.90 (m, 1H, C*H*HN), 4.58 (m, 1H, CH*H*N), 4.35 (m, 1H, C*H*), 4.03 (m, 1H, CHCH*H*), 3.41 (m, 1H, CHC*H*H), 2.79 (s, 5.3H, N(C$H_3$)$_3$), 2.65 (s, 3.7H, N(C$H_3$)$_3$); $^{13}C$ NMR ($CD_3OD$, 75.5 MHz) 173.04, 158.68, 158.50, 139.59, 137.74, 137.59, 130.12, 129.85, 129.78, 129.62, 129.53, 129.38, 129.16, 128.96, 128.84, 71.49, 70.33, 69.17, 68.91, 59.53, 59.03, 54.19, 53.76, 53.01; MS (CI) 371 ($MH^+$); Anal. Calc. for $C_{21}H_{26}N_2O_4$: C, 68.09; H, 7.07; N, 7.56. Found: C, 67.80; H, 7.03; H, 7.50.

**N-Benzyl-N-benzyloxycarbonyl-L-serine Methyl Ester (82)**

A solution of β-lactone **64** (97.3 mg, 0.313 mmol) in dry THF (10 mL) was added dropwise over 6 min to a solution of sodium methoxide (0.032 mmol) in dry methanol (5 mL). After 1 h, glacial acetic acid (0.02 mL) was added and the volume was reduced *in vacuo* . The resulting oil was partioned between water and $CHCl_3$. The organic phase was dried and concentrated *in vacuo* . The residue was purified by flash chromatography (80% hexanes/ethyl acetate; 50% hexanes/ethyl acetate) to give 95.1 mg (88%) of ester **82** as a colorless oil; $[\alpha]_D^{26}$ = -23°, c = 0.34 $CHCl_3$; IR ($CHCl_3$ cast) 3470, 3035, 2950, 1744, 1702, 1235, 701 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.28 (m, 10H, *Ph*), 5.28 (s, 2H, $CH_2OCO$), 4.61 (s, 2H, $CH_2N$), 4.5-3.8 (m, 3H, *CH*, $CH_2OH$), 3.77 (s, 3H, $OCH_3$), 3.65 (br s, 1H, OH); exact mass 343.1421, 284.1283 (343.1420, 284.1286 calc. for $C_{19}H_{21}NO_5$ and $C_{17}H_{18}NO_3$ respectively).

**N-Benzyl-N-benzyloxycarbonyl-L-α-aminobutyric Acid (83)**

Methyllithium (1.43 M in ether, 1.90 mL, 2.72 mmol) was added dropwise to a cooled slurry (-78 °C) of copper (I) bromide-dimethylsulfide complex (295 mg, 1.43 mmol) in dry THF (3 mL). This mixture was warmed to 0 °C, stirred for 20 min, and then cooled to -45 °C. A solution of β-lactone **64** (126 mg, 0.406 mmol) in dry THF (4 mL) was added dropwise over 8 min to the faintly yellow colored cuprate solution. The mixture was stirred 2.5 h at -45 °C and was then acidified with 1 N HCl. The organic solvents were removed *in vacuo* , the residue was filtered, and the filtrate was extracted with ethyl acetate (90 mL). The extracts were washed with brine (10 mL), dried, and concentrated *in vacuo* . The residue was purified by flash chromatography (chloroform; 95% chloroform/methanol) to afford 93.5 mg (70%) of **83**; $[\alpha]_D^{25}$ = -37°, c = 0.49, $CHCl_3$; IR ($CHCl_3$) 3100, 3030, 2965, 1705, 1247, 1081, 954, 698

$cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 10.38 (br s 1H, O*H*), 7.32 (s, 10H, *Ph*), 5.22 (s, 2H, $CH_2O$), 4.85-4.10 (m, 3H, C*H*, $CH_2N$) 1.92 (m, 2H, $CH_2CH_3$), 0.81 (t, J = 7 Hz, 3H, $CH_3$); exact mass 327.1469 (327.1471 calc. for $C_{19}H_{21}NO_4$).

**N-Benzyl-N-benzyloxycarbonyl-L-α-aminobutyric Acid (83) and (S)-N-Benzyl-N-benzyloxycarbonyl-3-amino-4-hydroxybutanone (84) using Cuprous Cyanide and Methyllithium**

Cuprous cyanide (0.105 g, 1.17 mmol) and dry THF (2 mL) were cooled to -78 °C under argon. Methyllithium (1.05 M in ether, 1.90 mL, 2.00 mmol) was added dropwise over 5 min and the mixture was allowed to warm slowly just until solution was complete.[125] The temperature was then brought to -78 °C and a solution of β-lactone **64** (0.204 g, 0.656 mmol) in THF (5 mL) was added dropwise over 5 min. The solution was stirred 90 min at -78 °C and 40 min at -45 °C before quenching with 1 N HCl. The organic solvents were removed *in vacuo* and the residue was filtered. The filtrate was extracted with ether (30 mL), dried, and concentrated *in vacuo* . The residue was purified by flash chromatography (50% hexanes/ethyl acetate; 99% ethyl acetate/acetic acid) to afford 10.5 mg (5%) of the ketone **84**, and a residue which was futher purified by ion exchange chromatography (Chelex-100, 50-100 mesh, $H^+$ form, 9 mL resin bed) to give 176.7 mg (82%) of acid **83**. The spectral and chromatographic properites of **83** were identical to those reported above.

For compound **84**: IR ($CHCl_3$) 3450, 3030, 2950, 1697, 1238, 1127, 700 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 200 MHz) δ 7.30 (m, 10H, *Ph*), 5.20 (m, 2H, Ph$CH_2O$), 4.56 (m, 2H, Ph$CH_2N$), 4.12 (m, 1H, C*H*), 3.8-3.4 (m, 2H, $CH_2O$), 3.26 (br s, 0.6H, O*H*), 2.36 (br s, 0.4H, O*H*), 2.00 (s, 1.8H, $CH_3$), 1.74 (s, 1.2H, $CH_3$); MS (CI) 328 ($MH^+$); Anal. Calc. for $C_{19}H_{21}NO_4$: C, 69.71; H, 6.47; N, 4.28. Found: C, 69.78; H, 6.42; N, 4.08.

**N-Benzyl-N-benzyloxycarbonyl-L-phenylalanine (85) from Phenylmagnesium Bromide**

A solution of bromobenzene (5.0 mL, 47 mmol) in dry THF (25 ml) was added dropwise to a suspension of Mg turnings (1.96 g, 80.6 mmol) in THF (50 mL). The mixture was stirred for 45 min and an aliquot was titrated with menthol and phenanthroline.[168] This Grignard reagent (0.527 M, 3.55 mL, 1.87 mmol) was added dropwise over 10 min to a cooled (-12 °C) suspension of copper (I) bromide-dimethylsulfide complex (197 mg, 0.957 mmol) and dimethyl sulfide (0.20 mL) in THF (5 mL) . The mixture was stirred for 2 h and a solution of β-lactone **64** (120 mg, 0.384 mmol) in THF (3 mL) was added dropwise. The pH was brought to 2 with 1 N $H_3PO_4$ after an additional 4 h. The resulting precipitate was filtered, the filtrate was concentrated *in vacuo* , and the residue was extracted with ethyl acetate (4 x 12 mL). The extracts were dried, concentrated *in vacuo* , and purified by flash chromatography (hexanes; 98% ethyl acetate/acetic acid) followed by reverse phase MPLC (60% $CH_3CN/H_2O$) to afford 24.6 mg (17%) of biphenyl and 89.5 mg (60%) of acid **85**.

For biphenyl: m.p. 68-70 °C (lit.[178] m.p. 69-71 °C); IR ($CHCl_3$) 3030, 1569, 1429, 1343, 902, 729, 697 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.52 (m, *Ph*); exact mass 154.0786 (154.0783 calc. for $C_{12}H_{10}$).

For acid **85**: $[\alpha]_D^{24}$ = -107, c = 0.59, $CHCl_3$; IR ($CHCl_3$) 3100, 3025, 2940, 1706, 1238, 1123, 986, 750, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 9.95 (br s, 1H, COO*H*), 7.25 (m, 15H, *Ph*), 5.26 (s, 2H, $CH_2O$), 4.7-3.72 (m, 3H, $CH_2N$, *CH*), 3.32 (br d, J = 6 Hz, 2H, $CH_2Ph$); exact mass 389.1631 (389.1627 calc. for $C_{24}H_{23}NO_4$); Anal. Calc. for $C_{24}H_{23}NO_4$: C, 74.02; H, 5.95; N, 3.60. Found: C, 74.11; H, 6.03; H, 3.36.

**N-Benzyl-N-benzyloxycarbonyl-L-phenylalanine (85) from Reaction of Phenyllithium and Cuprous Cyanide with 64**

The preparation of $Ph_2Cu(CN)Li_2$ followed the procedures outlined for $Me_2Cu(CN)Li_2$ above using CuCN (77.9 mg, 0.870 mmol) in THF (3 mL) and PhLi (2.14 M in 70% cyclohexane/ether, 0.70 mL, 1.5 mmol). The β-lactone **64** (151 mg, 0.485 mmol) in THF (6 mL) was added dropwise over 10 min to the cold (-78 °C) cuprate solution. The mixture was stirred for 1 h at -78 °C and the solution was allowed to warm to -15 °C over 3 h. It was then acidified, the organic solvents were removed *in vacuo*, and the residue was filtered. The filtrate was extracted with ether (35 mL), dried, and concentrated *in vacuo* . Purification by flash chromatography (90% hexanes/ethyl acetate; 99% ethyl acetate/acetic acid) afforded 47.9 mg (25%) of **85** as an oil. This material had spectral and chromatographic properties identical to those described above.

**N-Benzyl-N-benzyloxycarbonyl-L-α-aminoheptanoic Acid (86) and (S)-N-Benzyl-N-benzyloxycarbonyl-2-amino-1-hydroxyheptan-3-one (87)**

The cuprate, $(n\text{-Bu})_2Cu(CN)Li_2$, was formed from CuCN (0.101 g, 1.13 mmol) in THF (2.2 mL) and *n*-BuLi (1.2 M in hexanes/1.6 mL, 1.9 mmol) by a procedure analogous to that described above for $Me_2Cu(CN)Li_2$. A solution of β-lactone **64** (0.171 g, 0.548 mmol) in THF (3.3 mL) was added dropwise over 5 min to the cold (-78 °C) cuprate solution. The mixture was stirred for 40 min at -78 °C, warmed to -45 °C and allowed to slowly reach -36 °C over 1 h. The reaction mixture was acidified with 0.5 N HCl and filtered. The organic solvents were removed *in vacuo* from the filtrate and the residue was extracted with ether (40 mL). The ether layer was stirred with a saturated ethylenediaminetetraacetic acid (EDTA) solution (8 mL), washed with brine (10 mL), dried, and concentrated *in vacuo* to give an oil

which was partially purified by flash chromatography on silica gel (50% hexanes/ethyl acetate; 99% ethyl acetate/acetic acid). Fractions containing **86** and **87** were purified by reverse-phase MPLC (65% $CH_3CN/H_2O$) to give 154 mg (76%) of acid **86** and 11 mg (5%) of ketone **87**.

For acid **86**: $[\alpha]_D^{25}$ = -32°, c = 0.52, $CHCl_3$; IR ($CHCl_3$) 3100, 3030, 2956, 1706, 1235, 1100, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 200 MHz) δ 9.84 (br s, 1H, COO*H*), 7.26 (m, 10H, *Ph*), 5.18 (s, 2H, C$H_2$O), 4.64 (m, 1H), 4.5-4.15 (m, 2H, C$H_2$N), 1.91 (m, 1H), 1.74 (m, 1H), 1.11 (m, 6H, C$H_2$), 0.78 (m, 3H, $CH_3$) exact mass 369.1935 (369.1940 calc. for $C_{22}H_{27}NO_4$); Anal. Calc. for $C_{22}H_{27}NO_4$: C, 71.52; H, 7.37; N, 3.79. Found: C, 71.30; H, 7.43; N, 3.55.

For ketone **87**: IR ($CHCl_3$) 3440, 3030, 2955, 1700, 1233, 1125, 699 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 300 MHz) δ 7.34 (m, 10H, *Ph*), 5.20 (m, 2H, PhC$H_2$O), 4.53 (m, 2H, C$H_2$N), 4.11 (m, 1H, C*H*), 3.8 - 3.5 (m, 2H, C$H_2$O), 3.24 (m, 0.6H, O*H*), 2.25 (m, 2H, C$H_2$CO), 1.85 (m, 0.4H, O*H*), 1.40 (m, 1H, CH*H*), 1.17 (m, 2H, C$H_2$), 1.00 (m, 1H, C*H*H), 0.77 (m, 3H, C$H_3$); exact mass 369.1943, 284.1286 (369.1940 calc. for $C_{22}H_{27}NO_4$, 284.1286 calc. for $M^+$-$C_5H_9O$); Anal. Calc. for $C_{22}H_{27}NO_4$: C, 71.52; H, 7.37; N, 3.79. Found: C, 71.52; H, 7.28; N, 3.67.

**(S)-N-Benzyl-N-benzyloxycarbonyl-2-amino-4-methyl-hexanoic Acid (88) and Benzyl N-Benzyl Carbamate (89)**

The cuprate, (*sec*-Bu)$_2$Cu(CN)Li$_2$, was prepared as described for $Me_2Cu(CN)Li_2$ above from CuCN (83.1 mg, 0.928 mmol) in THF (2.3 mL) and *sec*-butyllithium (1.4 M in cyclohexane, 1.25 mL, 1.7 mmol). The β-lactone **64** (123 mg, 0.396 mmol) in THF (7 mL) was added dropwise over 4 min to the cold (-78 °C) cuprate solution. The solution was stirred for 20 min at -78 °C, 70 min at -45 °C, and 1 h at -18 °C before quenching with 1 N HCl. The organic solvents were removed *in*

*vacuo* . The residue was filtered and extracted with ether (35 mL). The extracts were dried, concentrated *in vacuo* to 5 mL, and washed with a saturated EDTA solution (3 mL). The ether phase was concentrated *in vacuo* and the residue was purified by chromatography on reverse-phase MPLC (70% $CH_3CN/H_2O$). Fractions containing **88** were purified further using flash chromatography (50% hexanes/ethyl acetate; 99% ethyl acetate/acetic acid) to afford 4.0 mg (4%) of **89** and 110 mg (76%) of pure **88** as an oil. For **89:** m.p. 59-61 °C (lit.[179] m.p. 60 °C); IR ($CHCl_3$) 3325, 1690, 1532, 1454, 1268, 1140, 748, 697 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 360 MHz) δ 7.34 (m, 10H, *Ph*), 5.15 (s, 2H, $CH_2O$), 5.10 (br s, 1H, N*H*), 4.38 (d, J = 6 Hz, 2H, $CH_2N$); exact mass 241.1103, (241.1102, calc. for $C_{15}H_{15}NO_2$).

For **88**: $[\alpha]_D^{28}$ = -38°, c = 0.47, $CHCl_3$; IR ($CHCl_3$) 3100, 3035, 2962, 1705, 1238, 1102, 975, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 400 MHz) δ 9.60 (br s, 1H, COO*H*), 7.26 (m, 10H, *Ph*), 5.18 (s, 2H, $CH_2O$), 4.7-4.4 (m, 3H, *CH*, $CH_2N$), 2.0-1.4 (m, 2H, $CH_2CHN$), 1.4-0.84 (m, 3H, $CHCH_3$, $CH_2CH_3$),0.84-0.59 (m, 6H, $CH_3$); exact mass 369.1938 (369.1940 calc. for $C_{22}H_{27}NO_4$); Anal. Calc. for $C_{22}H_{27}NO_4$: C, 71.52; H, 7.37; N, 3.79. Found: C, 71.32; H, 7.42; N, 3.70.

**Reaction of *tert*-Butyllithium with 64 to form (S)-N-Benzyl-N-benzyloxycarbonyl-2-amino-4,4-dimethylpentanoic Acid (90), Benzyl N-Benzyl Carbamate (89) and N-Benzyl-N-benzyloxycarbonyl-L-alanine (92)**

*Tert*-Butyllithium (2.0 mL, 3.0 mmol, 1.5 M in pentane) was added to a cooled (-78 °C) suspension of copper (I) bromide-dimethylsulfide complex (0.340 g, 1.66 mmol) in THF (3 mL). The mixture was stirred for 40 min at -78 °C and then for 20 min at -45 °C. A solution of β-lactone **64** (0.199 g, 0.382 mmol) in THF (3 mL) was added dropwise over 15 min. The mixture was stirred for 7 h, warmed to -10 °C over

1 h, and then was acidified with 1 N HCl and filtered. The filtrate was concentrated *in vacuo* and the residue was extracted with ethyl acetate (3 x 10 mL). The extracts were dried, concentrated *in vacuo*, and purified by flash chromatography (75% hexanes/ethyl acetate; 98% ethyl acetate/acetic acid). Fractions containing **90** and **92** were then purified using reverse-phase MPLC (60% $CH_3CN/H_2O$) to afford 71.9 mg (51%) of the desired acid **90**, 16.9 mg (18%) of urethane **89**, and 27.0 mg (23%) of alanine derivative **92**.

For **90**: m.p. 114-116 °C, $[\alpha]_D^{25}$ = -32.4°, c = 1.0, $CHCl_3$; IR ($CHCl_3$) 3100, 3030, 2957, 1706, 1367, 1244, 1120, 962, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 10.20 (br s, 1H, COO*H*), 7.30 (s, 10H, *Ph*), 5.22 (s, 2H, $CH_2O$), 4.74-4.27 (m, 3H, $CH_2N$, C*H*), 2.08 (dd, J = 5, 14 Hz, 1H, *t*-BuCH*H*), 1.60 (dd, J = 5, 14 Hz, 1H, *t*-BuC*H*H), 0.82 (s, 9H, $CH_3$); exact mass 369.1938 (369.1940 calc. for $C_{22}H_{27}NO_4$); Anal. Calc. for $C_{22}H_{27}NO_4$: C, 71.52; H, 7.37; N, 3.79. Found: C, 71.25; H, 7.31; N, 3.74.

Compound **89** possessed spectral and chromatographic properties identical to those described above.

For **92**: $[\alpha]_D^{25}$ = -29°, c = 0.38, $CHCl_3$; IR ($CHCl_3$) 3100, 3030, 2942, 1704, 1260, 1213, 1070, 1015, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 8.75 (br s, 1H, COO*H*), 7.27 (s, 10H, *Ph*), 5.22 (s, 2H, $CH_2O$), 4.9-4.2 (m, 3H, $CH_2N$, C*H*), 1.37 (d, J = 7 Hz, 3H, $CH_3$); exact mass 313.1311 (313.1314 calc. for $C_{18}H_{19}NO_4$).

**(S)-N-Benzyl-N-benzyloxycarbonyl-2-amino-4,4-dimethylpentanoic Acid (90), Benzyl N-Benzyl Carbamate (89) and N-Benzyl-N-**

**benzyloxycarbonyl-L-alanine (92) from Reaction of Cuprous Cyanide and *tert*-Butyllithium with 64**

The cuprate, $(t\text{-Bu})_2Cu(CN)Li_2$, was prepared as described above for $Me_2Cu(CN)Li_2$ from CuCN (68.7 mg, 0.767 mmol), THF (2 mL) and *t*-BuLi (1.38 M in pentane, 1.0 mL, 1.38 mmol). The β-lactone **64** (126 mg, 0.404 mmol) in THF (2 mL), was introduced dropwise to the cold (-78 °C) cuprate solution. This mixture was stirred for 1 h at -78 °C, 1 h at -45 °C, and 30 min at -15 °C. The mixture was acidified with 1 N HCl and concentrated *in vacuo* . The residue was filtered and the filtrate was extracted with ether (4 x 7 mL). The ether extract was dried, concentrated *in vacuo* and purified by flash chromatography (70% hexanes/ethyl acetate; 99% ethyl acetate/acetic acid) to afford 56.3 mg (38%) of **90**, 18.4 mg (19%) of **89** and 18 mg (14%) of impure **92**. The chromatographic and spectral properties of these compounds were identical to those described above.

**(S)-Methyl N-Benzyl-N-benzyloxycarbonyl-2-aminobutanoate (95)**

To a crude sample of **83** (79.5 mg, 0.243 mmol) in ether (5 mL) was added an etheral solution of diazomethane until a yellow color persisted. Evaporation of the solvent followed by flash chromatography (80% hexanes/ethyl acetate) afforded 67.0 mg (84%) of ester **95** as an oil; $[\alpha]_D^{24}$ = -49.7°, c = 0.59, $CHCl_3$; IR ($CHCl_3$) 3025, 2942, 1742,1703, 1250, 1210, 1135, 1080, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 200 MHz) δ 7.28 (m, 10H, *Ph*), 5.19 (s, 2H, $CH_2O$), 4.56 (m, 2.6H, $CH_2N$, *CH*), 4.14 (m, 0.4 H, *CH*), 3.56 (s, 1.6H, $OCH_3$), 3.41 (s, 1.4H, $OCH_3$), 1.97 (m, 1H, $CHHCH_3$), 1.78 (m, 1H, $CHHCH_3$), 0.83 (m, 3H, $CH_3$); exact mass 341.1623 (341.1627 calc. for $C_{20}H_{23}NO_4$); Anal. Calc. for $C_{20}H_{23}NO_4$: C, 70.36; H, 6.79; H, 4.10. Found: C, 70.66; H, 6.94; N, 4.10.

**Reaction of 64 with Organocerium Reagents to give (S)-N-Benzyl-N-benzyloxycarbonyl-2-amino-4-hydroxybutanone (84) and (S)-N-Benzyl-N-benzyloxycarbonyl-2-amino-3-methylbutan-1,3-diol (96)**

The organocerium reagent was prepared according to the procedure of Imamoto *et al.*[152] Commerical cerium chloride heptahydrate (0.155 g, 0.416 mmol) was dried at 135 °C *in vacuo* (0.05 mmHg) for 2 h. Upon cooling to room temperature, with stirring under Ar, THF (5 mL) was added. The resulting suspension was stirred for 2 h and then cooled to -78 °C. Methyllithium (1.2 M in ether, 0.35 mL, 0.42 mmol) was added dropwise. The yellow mixture was stirred for 35 min and a solution of β-lactone **64** (98.5 mg, 0.316 mmol) in THF (3 mL) was added dropwise. The mixture was warmed after 3.5 h to 20 °C and acidified to pH 1.5 with 1 N HCl. The mixture was concentrated *in vacuo*. The residue was extracted with ether (3 x 8 mL), dried, concentrated *in vacuo*, and purified by flash chromatography (65% hexanes/ethyl acetate; ethyl acetate) to afford 55.9 mg (57%) of recovered starting material (**64**), 11.3 mg (11%) of **84** and 20.4 mg (19%) of **96**.

Compound **84** possessed spectral and chromatographic properties identical to those desribed above.

For **96**: $[\alpha]_D$ = -29°, c = 0.20, $CHCl_3$; IR ($CHCl_3$) 3410, 2970, 1672, 1499, 1477, 1347, 1236, 1144, 1116, 1050, 698 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 200 MHz) δ 7.35 (m, 10H, *Ph*), 5.29 (d, J = 12 Hz, 1H, PhCH*H*O), 5.21 (d, J = 12 Hz, 1H, PhC*H*HO), 4.66 (d, J = 14 Hz, 1H, PhCH*H*N), 4.58 (d, J = 14 Hz, 1H, PhC*H*HN), 4.05 (m, 2H, CHC$H_2$), 3.32 (br s, 0.83H, O*H*), 2.05 (br s, 0.83H, O*H*) 1.79 (s, 0.34H, O*H*), 1.24 (s, 3H, C$H_3$) 0.97 (s, 3H, C$H_3$); MS (CI) 344 ($MH^+$); Anal. Calc. for $C_{20}H_{25}NO_4$: C, 69.95; H, 7.34; N, 4.08. Found: C, 69.95; H, 7.39; N, 3.75.

**(S)-2-Aminobutanoic Acid (97)**

A suspension of 10% palladium on charcoal (0.189 g) in isopropyl alcohol (7 mL) was prehydrogenated for 35 min at atmospheric pressure. To this was added a solution of **83** (0.127 g, 0.388 mmol), isopropyl alcohol (6 mL), ethyl ether (3 mL) and 1 N HCl (0.80 mL). The mixture was stirred under one atmosphere of hydrogen for 9 h, and was filtered through a Celite pad. The solvent was removed *in vacuo* and an aqueous solution of the residue was applied to an ion exchange column (AG50W-X8, 50-100 mesh, $H^+$ form, 13 mL bed volume). Elution with 2% aqueous ammonia followed by lyophilization gave 39.6 mg (99%) of **97**, m.p. 260 (dec.), $[\alpha]_D^{25}$ = +6.4°, c = 1.0, $H_2O$ (lit.[178] m.p. 270-280 °C, $[\alpha]_D$ = -7.94°, c = 4, $H_2O$, (*D*),[171] $[\alpha]_D^{25}$ = +7.7°, c = 1.0, $H_2O$[180]); IR (KBr) 3020, 2975, 1605, 1584, 1517, 1407 $cm^{-1}$; $^1H$ NMR ($D_2O$, 300 MHz) δ 3.72 (t, J = 5.5 Hz, 1H, C*H*), 1.92 (m, 2H, $CH_2$), 1.00 (t, J = 7.5 Hz, 3H, $CH_3$); MS (FAB) 104 ($MH^+$).

**L-Phenylalanine (98)**

A mixture of 85 mg of 10% palladium on charcoal in isopropyl alcohol (3 mL) was prehydrogenated for 2 h at atmospheric pressure. To this was added a solution of **85** (36 mg, 0.092 mmol), isopropyl alcohol (5 mL), ethyl ether (1 mL), and 1 N HCl (0.21 mL). The mixture was stirred for 7 h under one atmosphere of hydrogen and was then filtered. The solvent was removed *in vacuo* . Ion exchange chromatography (15 mL bed volume) and lyophilization, as described for **97** above, afforded 12 mg (79%) of **98** , with chromatographic properties identical to authentic L-phenylalanine, m.p. 250 °C (dec.) (lit.[171] m.p. 270-275 °C (dec.); $[\alpha]_D^{25}$ = -22.0°, c = 0.49, $H_2O$ (lit.[178] $[\alpha]_D$ = -35.1°, c = 1.94, $H_2O$); IR (KBr) 3023, 3000, 1612, 1586, 1500, 1402, 742, 698 $cm^{-1}$; $^1H$ NMR ($D_2O$, 400 MHz) δ 7.40 (m, 5H, *Ph*), 4.00 (dd, J = 5.0, 8.0 Hz, 1H, C*H*), 3.30 (dd, J = 5.0, 14.5 Hz, 1H, C*H*H), 3.13 (dd, J = 8.0,

14.5 Hz, 1H, CH*H*); $^{13}C$ NMR ($D_2O$, 100.6 MHz) δ 175.0, 136.5, 130.7, 130.4, 128.8, 56.4, 36.7; MS (FAB) 166 ($MH^+$).

**(S)-2-Amino-4,4-dimethylpentanoic Acid (99)**

A solution of **90** (0.158 g, 0.428 mmol), isopropyl alcohol (6 mL), ethyl ether (3 mL), and 1 N HCl (0.9 mL) was added to 0.36 g of 10% palladium on charcoal which had been prehydrogenated for 1 h in isopropyl alcohol (7 mL). The mixture was stirred under 1 atm of hydrogen for 10 h, filtered, and the solvent was removed *in vacuo* . The residue was applied as a water/methyl alcohol (9:1) solution to an ion exchange column (AG50W-X8, 50-100 mesh, $H^+$ form, 12 mL bed volume)and eluted with 2% aqueous ammonia followed by lyophilization to give 62 mg (quantitative yield) of **99**, m.p. 254 °C (dec.), $[\alpha]_D^{25}$ = -11.3°, c = 1, $H_2O$, $[\alpha]_D^{25}$ = +15.6°, c = 1.1, glacial acetic acid (lit.[4] m.p. 260 °C, $[\alpha]_D^{23}$ = -12.6°, c = 1, $H_2O$, $[\alpha]_D$ = +14.7°, acetic acid,[181] $[\alpha]_D^{25}$ = +16.3, c = 1.2, glacial acetic acid[182]); IR (KBr) 3000, 2957, 2140, 1658, 1622, 1577, 1515, 1410, 1276, 1153 $cm^{-1}$; $^1H$ NMR ($CD_3OD$, 400 MHz) δ 3.43 (dd, J = 4.6, 7.3 Hz, 1H, C*H*); 1.95 (dd, J = 4.6, 14.6 Hz, 1H, C*H*H), 1.45 (dd, J = 7.3, 14.6 Hz, 1H, CH*H*), 0.89 (s, 9H, $CH_3$); MS (FAB) 146 ($MH^+$).

**Preparation of (2'R)-Methyl ([(1S,4R)-4,7,7,-Trimethyl-3-oxo-2-oxabicyclo[2.2.1.]heptane-1-carbonyl]amino)butanoate (103) and Derivatives 100, 102, and 104**

The procedure of Armarego *et al*[155] was adopted. In a typical experiment, D-α aminobutyric acid (**106**) (0.206 g, 2.00 mmol) in 2 M NaOH (1 mL) was added to a cooled (0 °C) solution of (-)-camphanoyl chloride (0.651 g, 3.00 mmol) in toluene (1.5 mL) followed by 1 mL of 3 M NaOH. The mixture was stirred at room temperature for 3 h with periodic addition of 2 M NaOH to keep the pH above 7. The solution was

washed with dichloromethane (5 mL). The aqueous phase was acidified to pH 1 with 1 M HCl, and was then extracted with dichloromethane (4 × 5 mL). These extracts were dried and concentrated *in vacuo* . The residue was dissolved in ether (12 mL) and an etheral solution of diazomethane was added until a yellow color persisted. Evaporation of the solvent *in vacuo* and purification of the residue by flash chromatography (80% hexanes/ethyl acetate; 60% hexanes/ethyl acetate) gave 0.508 g, (86%) of **103** as a colorless oil. The (2'S) and (2'RS) isomers, **102** and **104,** were prepared similarly.

For **103**: $[\alpha]_D^{25}$ = -13.8, c = 1.06, $CHCl_3$; IR ($CHCl_3$) 3415, 3360, 2960, 1793, 1745, 1676, 1527, 1267, 1212, 1178, 1169, 1125, 1061 $cm^{-1}$; $^1H$ NMR ($C_6D_6$, 300 MHz) δ 6.75 (br d, J = 7.5 Hz, 1H, N*H*), 4.59 (m, 1H, C*H*), 3.22 (s, 3H, OC$H_3$), 2.35 (m, 1H, *H*-6' endo), 1.64 (m, 2H, C$H_2$), 1.4-1.15 (m, 3H, C$H_2$, *H*-6' exo), 0.99 (s, 3H, C$H_3$), 0.85 (s, 6H, C$H_3$), 0.66 (t, J = 7.5 Hz, 3H, $CH_2CH_3$); $^{13}C$ NMR ($C_6D_6$, 75.5 MHz) δ 177.30, 171.98, 167.26, 92.14, 55.21, 53.96, 53.34, 51.60, 30.61, 28.97, 24.99, 16.57, 16.53, 9.90, 9.75; exact mass 297.1580 (297.1576 calc. for $C_{15}H_{23}NO_5$); Anal. Calc. for $C_{15}H_{23}NO_5$: C, 60.59; H, 7.80; N, 4.71. Found: C, 60.69; H, 7.78; N, 4.72.

For **102** prepared from authentic L-α-aminobutyric acid (**105**): m.p. 74-76 °C; $[\alpha]_D^{25}$ = -16.5, c = 1.08, $CHCl_3$. The spectral properties are consistent with those reported above.

For **104** from authentic DL-α-aminobutyric acid (**107**) the spectral properties are consistent with those reported above.

A sample of amino acid **97**, obtained from hydrogenation of diprotected phenylalanine derivative **83**, was treated in the same manner to afford a crude product containing the camphanamide methyl ester **100**. This compound was not purified by silica gel chromatography but was directly analyzed by gas chromatography (150 °C 1 min, 5 °C/min to 240 °C, 8.5 psi). Table 3 summarizes the results.

**N-[(1S,4R)-(4,7,7-Trimethyl-3-oxo-2-oxabicyclo[2.2.1.]heptane-1-carbonyl]-phenylalanine Methyl Ester (108) and 2'S Isomer 101**

The procedure of Armarego *et al*[155] was followed as described for **103** above to acylate a mixture of D-phenylalanine (0.209 g, 1.27 mmol) and L-phenylalanine (0.100 g, 0.61 mmol) with (-)-camphanoyl chloride (0.600 g, 2.77 mmol) followed by esterification with diazomethane. Purification by flash chromatography (90% hexanes/ethyl acetate; 60% hexanes/ethyl acetate) gave 0.519 g (77%) of **108**; IR ($CHCl_3$) 3400, 3360, 3025, 2960, 1789, 1745, 1675, 1526, 1265, 1219, 1176, 1168, 1122, 1059, 747, 699 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 300 MHz) δ 7.27 (m, 3H, *Ph*), 7.15 (m, 2H, *Ph*), 6.81 (br d, J = 8.5 Hz, 1H, N*H*), 4.91 (m, 1H, C*H*), 3.73 and 3.72 (2s, 3H, OC$H_3$), 3.22 (dd, J = 5.5, 14 Hz, 1H, CH*H*Ph), 3.04 (m, 1H, C*H*HPh), 2.42 (m, 1H, *H*-6' endo), 1.97-1.6 (m, 3H, C$H_2$, *H*-6' exo), 1.09, 1.07, 1.06 (3s, 5H, C$H_3$), 1.01 (s, 1H, C$H_3$), 0.88 (s, 2H, C$H_3$), 0.61 (s, 1H, C$H_3$); exact mass 359.1729 (359.1733 calc. for $C_{20}H_{25}NO_5$); Anal. Calc. for $C_{20}H_{25}NO_5$: C, 66.83; H, 7.01; N, 3.90. Found: C, 66.47; H, 7.11; N, 3.89.

A sample of amino acid **98** obtained from hydrogenation of **85** was treated in the same manner to afford a sample of the 2'S isomer **101**; $^1H$ NMR ($CDCl_3$, 300 MHz) δ 7.27 (m, 3H, *Ph*), 7.15 (m, 2H, *Ph*), 6.81 (d, J = 8.5 Hz, 1H, N*H*), 4.95 (dt, J = 5.5, 8.5 Hz,1H, C*H*), 3.73 (s, 3H, OC$H_3$), 3.22 (dd, J = 5.5, 14 Hz, 1H, C*H*HPh), 3.01 (dd, J = 8.5, 14 Hz, 1H, CH*H*Ph) 2.45 (m, 1H, *H*-6' endo), 1.90 (m, 2H, C*H*H), 1.66 (m, 1H, CH*H*), 1.07 (s, 3H, C$H_3$), 1.01 (s, 3H, C$H_3$), 0.61 (s, 3H, C$H_3$). The IR and mass spectral data are identical to those reported above. The stereochemical purity of this compound was determined by gas chromatography (170 °C, 5 °C/min to 240 °C, 240 °C 10 min, 8.5 psi). The results are summarized in Table 4.

**DL-2-(Tetrahydro-2,6-dioxo-2H-pyran-3-yl)-1H-isoindole-1,3(2H)-dione (109)**

The procedure of Lindsay and Whitaker was used.[159] Phthalic anhydride (48.2 g, 0.326 mol) and L-glutamic acid (48.0 g, 0.326 mol) were mixed and heated to 180 °C. After 30 min the mixture was cooled to 100 °C and acetic anhydride (65.0 g, 0.645 mol) was added. Xylene (185 mL) was added, and after 5 min the mixture was cooled to 4 °C and filtered to give 39.9 g (47%) of anhydride **109**, m.p. 201-203 °C, (lit.[159] m.p. 199-201 °C); IR (KBr) 3060, 2915, 1816, 1777, 1767, 1715, 1470, 1390, 720 $cm^{-1}$; $^1H$ NMR ($Me_2SO$-$d_6$, 80 MHz) δ 7.95 (m, 4H, *Ar*) 5.49 (dd, J = 6.0, 15 Hz, IH, C*H*), 3.04 (m, 2H, C$H_2$CO), 2.64 (m, 1H, CHCH*H*), 2.25 (m, 1H, CHC*H*H); exact mass 277.0479 (277.0481 calc. for $C_{13}H_9NO_5$); Anal. Calc. for $C_{13}H_9NO_5$: C, 60.24; H, 3.50, N, 5.40. Found: C, 59.90; H, 3.56; N, 5.57.

**Thalidomide (36)**

The procedure of Helm *et al*[97] was used. Finely ground urea (2.20 g, 36.6 mmol) and phthaloylglutamic anhydride **109** (18.6 g, 71.8 mmol) were mixed and then heated to 180 °C for 1 h. The mixture was cooled and repeatedly recrystallized from glacial acetic acid to give 7.36 g (40%) of **36**, m.p. 267-269 °C (lit.[97] m.p. 269-271 °C); IR (KBr) 3195, 3095, 2955, 1786, 1773, 1733, 1710, 1696, 1470, 1387, 729 $cm^{-1}$; $^1H$ NMR ($Me_2$ SO-$d_6$, 200 MHz) δ 11.21 (s, 1H, N*H*), 7.94 (m, 4H, *Ar*), 5.22 (dd, J = 5.5, 12.5 Hz, 1H, C*H*), 2.95 (m, 1H, C*H*C$H_2$CO), 2.62 (m, 2H, C$H_2$CO), 2.12 (m, 1H, C*H*C$H_2$CO); $^{13}C$ NMR ($Me_2SO$-$d_6$, 22.6 MHz) δ 172.7, 169.8, 167.1, 134.9, 131.2,123.4, 49.0, 30.9, 22.0; exact mass 258.0640 (258.0640 calc. for

$C_{13}H_{10}N_2O_4$); Anal. Calc. for $C_{13}H_{10}N_2O_4$: C, 60.47; H, 3.90; N, 10.85. Found: C, 60.49; H, 3.98; N, 10.79.

**N-Phthaloyl-L-glutamic acid (110) from 109**

The procedure of King and Kidd[160] was used. Anhydride **109** (40.0 g, 0.154 mol) was added to 150 mL of boiling water. The solution was heated for 10 min and was then slowly cooled to 4 °C. The resulting white crystals were filtered and washed with ice cold water to give 40.4 g (94%) of diacid **109**, m.p. 162-166 °C, $[\alpha]_D^{25}$ = -37.7 °, c = 1, 95% ethanol (lit.[161] m.p. 158-160 °C, $[\alpha]_D^{25}$ = -46.1°, c = 1, 95% ethanol); IR (KBr) 3000, 1776, 1734,1714, 711 $cm^{-1}$; $^1H$ NMR (acetone-$d_6$, 80 MHz) δ 8.97 (br s, 2H, COO*H*), 7.90 (s, 4H, *Ar*), 5.00 (m, 1H, C*H*), 2.47 (m, 4H, $CH_2CH_2COO$); exact mass 277.0585 (277.0586 calc for $C_{13}H_{11}NO_6$); Anal. Calc. for $C_{13}H_{11}NO_6$:C, 56.32; H, 4.00; N, 5.05. Found: C, 56.34; H, 3.91; N, 5.06.

**Preparation of 110 from Glutamic Acid**

The procedure of Elberling *et al*[161] was followed. Solid N-carboethoxyphthalimide (25.0 g, 0.114 mol) was added slowly in portions to a chilled ( 3 °C) solution of L-glutamic acid (16.8 g, 0.114 mol) in 235 mL of 1.06 M aqueous sodium carbonate (0.25 mol.) When the addition was complete, the mixture was allowed to warm to 20 °C overnight. The precipitate was filtered, cooled to 0 °C, and acidified with concentrated hydrochloric acid. The resulting oil crystallized overnight at 4 °C. Recrystallization from water gave 12.6 g (40%) of **110**, m.p. 158-160 °C, $[\alpha]_D^{25}$ = -45.7°, c = 1.0, 95% ethanol (lit.[161] m.p. 158-160 °C, $[\alpha]_D^{26}$ = -46.1° c = 1, 95% ethanol). The spectral properties were identical to those of **110** obtained from **109**.

**2-(1,3-Dihydro-1-oxo-2H-isoindol-2-yl)-pentanedioic Acid (111)**

A modification of the procedure of Jönsson *et al*[183] was employed. Zinc dust (39.4 g, 0.603 mol) was added to diacid **110** (31.6 g, 0.114 mol) in 280 mL of boiling glacial acetic acid. The mixture was heated to reflux for 4 h and filtered hot. The precipitate was washed with acetic acid, and the solvent was removed *in vacuo*. Recrystallization from water and washing with $CHCl_3$ afforded 18.8 g (63%) of phthalimidino diacid **111**, m.p. 219 °C (dec.) (lit.[183] m.p. 220 °C); IR (KBr) 2980, 1744, 1713, 1644, 1475, 1459, 1430, 743 $cm^{-1}$; $^1H$ NMR ($Me_2$ SO-$d_6$, 80 MHz) δ 12.62 (br s, 2H, COO*H*), 7.68 (m, 4H, *Ar*), 4.90 (m, 1H, C*H*), 4.55 (s, 2H, C$H_2$N), 2.32 (m, 4H, C$H_2$C$H_2$COO); exact mass 263.0794 (263.0794 calc. for $C_{13}H_{13}NO_5$); Anal. Calc. for $C_{13}H_{13}NO_5$: C, 59.31; H, 4.98; N, 5.32. Found: C, 59.10; H, 4.93; N, 5.31.

**2-(Tetrahydro-2,6-dioxo-2H-pyran-3-yl)-1H-isoindole-1-one (112)**

The procedure of Jönsson *et al*[183] was followed. Acetic anhydride (5.0 mL, 53 mmol) was added to diacid **111** (1.04 g, 3.95 mmol) and the mixture was heated to reflux for 5 min. Cooling produced crystals which were filtered and washed with $CHCl_3$ to give 0.931 g (96%) of phthalimidino anhydride **112**, m.p. 250-253 °C (lit.[184] m.p. 254-257 °C); IR (KBr) 2908, 1813, 1777, 1676, 1473, 1453, 734 $cm^{-1}$; $^1H$ NMR ($Me_2$ SO-$d_6$, 80 MHz) δ 7.60 (m, 4H, *Ar*), 5.41 (dd, J = 6, 13 Hz, 1H, C*H*), 4.47 (s, 2H, ArC$H_2$N), 3.02 (m, 2H, C$H_2$CO), 2.55 (m, 1H, CHC*H*H), 2.15 (m, 1H, CHCH*H*); exact mass 245.0689 (245.0688 calc. for $C_{13}H_{11}NO_4$); Anal. Calc. for $C_{13}H_{11}NO_4$: C, 63.67; H, 4.52; N, 5.71. Found: C, 63.40; H, 4.54; N, 5.74.

**2-(2,6-Dioxo-3-piperidinyl)-1H-isoindole-1-one (37)**

The procedure of Helm *et al*[97] was followed. Urea (0.127 g, 2.12 mmol) and **112** (1.01 g, 4.12 mmol) were mixed and heated to 180 °C for 40 min. The mixture was cooled, recrystallized from glacial acetic acid and from *n*-butanol to afford 0.407 g (40%) of **37**, m.p. 234-236 °C (lit.[97] 238-240 °C); IR (KBr) 3200, 3095, 2915, 1732, 1704, 1680, 1378, 731 $cm^{-1}$; $^1H$ NMR (dioxane-$d_8$, 200 MHz) δ 9.93 (s, 1H, N*H*), 7.83 (m, 1H, *Ar*), 7.52 (m, 3H, *Ar*), 5.22 (dd, J = 5.0, 13 Hz, 1H, C*H*), 4.40 (d, J = 16 Hz, 1H, CH*H*N), 4.32 (d, J = 16 Hz, 1H, C*H*HN), 2.74 (m, 2H, $CH_2$CO), 2.28 (m, 1H, CHC*H*H), 2.08 (m, 1H, CHCH*H*); exact mass 244.0847 (244.0847 calc. for $C_{13}H_{12}N_2O_3$); Anal. Calc. for $C_{13}H_{12}N_2O_3$: C, 63.93; H, 4.95; N, 11.47. Found: C, 63.84; H, 4.93; N, 11.54.

**2-(Bromomethyl)benzoic Acid (114)**

Compound **114** was prepared following the procedure of Pifferi and Testa.[163] A solution of 25.0 g (0.186 mol) of phthalide (**113**) in glacial acetic acid (70 mL) was added to 170 mL of glacial acetic acid which had been saturated with HBr gas at 5 °C. More HBr gas was added, the ice bath was removed and the mixture was stirred 4 h at 20 °C and 1 h at 80 °C. The mixture was cooled overnight and the precipitate was collected and washed with small volumes of ice cold water. A second crop of crystals was obtained by pouring the filtrate onto 400 g of ice and filtering. The solid was dried to give 23.9 g (60%) of **114**, m.p. 145-146 °C (lit.[163] m.p. 149-150 °C); IR (THF) 2900, 1679, 1405, 701 $cm^{-1}$; $^1H$ NMR (acetone-$d_6$, 100 MHz) δ 8.04 (m, 1H, *Ar*), 7.58 (m, 3H, *Ar*), 5.09 (s, 2H, $CH_2$); exact mass 215.9608, 213.9632 (215.9610, 213.9630 calc. for $C_8H_7BrO_2$); Anal. Calc. for $C_8H_7BrO_2$: C, 44.68; H, 3.28. Found: C, 44.75; H, 3.32.

**2-(Bromomethyl)benzoyl Chloride (115)**

The procedure of Pifferi and Testa[163] was followed with slight modification. Thionyl chloride (40.0 mL, 0.551 mol) and bromo acid **114** (37.5 g, 0.174 mol) were heated to reflux for 5 h. -The excess thionyl chloride was removed *in vacuo* . Dry benzene (30 mL) was added, and the solvent was removed *in vacuo* . Several repetitions provided a crystalline mass which was distilled to afford 38.3 g (94%) of acid chloride **115**, b.p. 74-79 °C (0.03 mm Hg), m.p. 45-47 °C (lit.[163] m.p. 46-48 °C; IR ($CCl_4$) 1766, 1596, 1570, 1478, 1448, 1188, 857, 768 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 8.28 (m, 1H, *Ar*), 7.57 (m, 3H, *Ar*), 4.79 (s, 2H, $CH_2$); exact mass 235.9247, 233.9271, 231.9295 (235.9242, 233.9271, 233.9261, 231.9291 calc. for $C_8H_6BrClO$); Anal. Calc. for $C_8H_6BrClO$: C, 41.15; H, 2.59. Found: C, 41.41; H, 2.60.

**Acetohydrazide (116)**

The procedure of Curtius and Hofman was followed.[164] Ethyl acetate (850 mL, 8.62 mol) was added to hydrazine hydrate (500 mL, 10.3 mol) at 0 °C. This mixture was heated to reflux for 48 h and cooled to 4 °C overnight. The solid was filtered and dried to give 318 g (50%) of acetohydrazide (**116**), m.p. 60-67 °C (lit.[171] m.p. 62-67 °C); IR ($CHCl_3$) 3280, 3045, 1665, 1637, 1531, 1374, 1310 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 8.15 (br s, 1H, N*H*), 4.03 (br s, 2H, $NH_2$), 1.99 (s, 3H, $CH_3$); exact mass 74.0480 (74.0485 calc. for $C_2H_6N_2O$).

**N-(1,3-Dihydro-1-oxo-2H-isoindol-2-yl)-acetamide (117)**

A modification of the procedure of Bellasio and Testa[112] was used. Triethylamine (30.0 mL, 0.215 mol) was added to a solution of 15.7 g (0.212 mol) of acetohydrazide (**116**) in dry dioxane (45 mL). A solution of acid chloride **114** (20.7

g, 88.6 mmol) in dry dioxane (70 mL) was added dropwise and the temperature was maintained below 20 °C. The solution was then refluxed for 2 h and filtered. The filtrate was concentrated *in vacuo* and washed with aqueous HCl (pH 3.5) to afford 10.7 g (63%) of **117**, m.p. 198-199 °C (lit.[112] m.p. 196-200 °C); IR ($CHCl_3$) 3200, 3000, 1710, 1672, 1550, 770 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 100 MHz) δ 9.72 (s, 1H, N*H*) 7.85 (m, 1H, *Ar*), 7.50 (m, 3H, *Ar*), 4.69 (s, 2H, C$H_2$), 2.13 (s, 3H, COC$H_3$); exact mass 190.0745 (190.0743 calc. for $C_{10}H_{10}N_2O_2$): Anal. Calc. for $C_{10}H_{10}N_2O_2$: C, 63.15; H, 5.30; N, 14.73. Found: C, 63.13; H, 5.32; N, 14.51.

**Preparation of the Hydrochloride Salt of 118 from 117**

The HCl salt of hydrazide **118** was prepared using the procedure of Bellasio and Testa.[112] Compound **117** (5.41 g, 28.4 mmol) was heated to reflux for 5 min with concentrated HCl (10 mL), dioxane (35 mL) was added, the solution was cooled slowly, and the resulting crystals were filtered and washed with $CHCl_3$ to give 3.50 g (67%) of the HCl salt of **118**, m.p. 198-201 °C (lit.[112] m.p. 202-205 °C); IR (MeOH) 2700, 1719, 1618, 1530, 738 $cm^{-1}$; $^1H$ NMR ($Me_2SO$-$d_6$, 360 MHz) δ 10.85 (br s, 3H, N$H_3$), 7.84-7.50 (m, 4H, *Ar*), 4.80 (s, 2H, C$H_2$); MS (FAB) 185 ($M^+$ + HCl), 149 ($MH^+$); Anal. Calc. for $C_8H_9N_2ClO$: C, 52.04; H, 4.91; N, 15.17. Found: C, 51.76; H, 4.88; N, 14.99.

**Phthalazone (120)**

A modification of the procedure of Gabriel and Neumann[166] was followed. Hydrazine hydrate (13.6 mL, 0.280 mol) was added dropwise to a solution of *o*-formylbenzoic acid (40.9 g, 0.272 mol) in 160 mL of 98% ethanol. The solution turned yellow and a white solid precipitated which was filtered and washed with 98% ethanol to give 35.6 g (90%) of **120**, m.p. 183.5-185 °C (lit.[166] m.p. 182 °C); IR

($CHCl_3$) 3160, 1663, 1350, 1153, 771 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 11.41 (br s, 1H, N*H*) 8.55 (m, 1H, *Ar*), 8.25 (s, 1H, NC*H*), 7.85 (m, 3H, *Ar*); exact mass 146.0480 (146.0480 calc. for $C_8H_6N_2O$); Anal. Calc. for $C_8H_6N_2O$: C, 65.75; H, 4.14; N, 19.17. Found: C, 65.53; H, 4.11; H, 19.19.

**2-Amino-2,3-dihydro-1H-isoindol-1-one (118) from 120**

The procedure of Darapsky and Heinrichs[165] was used with slight modification. Zn dust (62.0 g, 0.948 mol) was added to a solution of phthalazone **120**, (22.4 g, 0.153 mol) in 100 mL of 2 N NaOH. An additional 175 mL of 2 N NaOH was added and the mixture was heated to reflux for 3 h. After cooling the excess zinc dust was filtered, and the filtrate was extracted with $CHCl_3$ in a continuous extractor. The $CHCl_3$ phase was dried, filtered, and concentrated *in vacuo* to give 16.0 g (71%) of hydrazide **118** as a white solid which was used without further purification; IR ($CHCl_3$) 3300, 3190, 1690, 1620, 732 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.80 (m, 1H, *Ar*), 7.47 (m, 3H, *Ar*), 4.45 (br s, 4H, $CH_2$, $NH_2$); exact mass 148.0636 (148.0637 calc. for $C_8H_8N_2O$).

**N-Phthalimidinophthalimidine (45)**

The procedure of Bellasio and Testa[112] was followed. A solution of acid chloride **115** (4.27 g, 18.3 mmol) in dry dioxane (25 mL) was added dropwise to a cooled (10 °C) solution of hydrazide **118** (2.70 g, 18.2 mmol) in dry dioxane (50 mL) at a rate to maintain the temperature at 10-13 °C. A solution of triethylamine (5.6 mL, 4.1 g, 40 mmol) in dioxane (10 mL) was then added dropwise at 10-13 °C. After the addition was complete, the mixture was heated to 60 °C for 3.5 h. The suspension was filtered, and the filtrate was concentrated *in vacuo* . The residue was washed with water, recrystallized from dioxane, washed with small volumes of chloroform, and

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recrystallized from chloroform/cyclohexane to give 1.77 g (37%) of **45**, m.p. 282-284 °C (lit.[112] m.p. 283-285 °C); IR ($CHCl_3$) 3060, 3040, 3020, 1717, 1699, 1688, 1658, 1372, 734 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 200 MHz) δ 7.96 (d, J = 6 Hz, 2H, *Ar*), 7.60 (m, 6H, *Ar*), 4.83 (s, 2H, $CH_2$); exact mass 264.0899 (264.0899 calc. for $C_{16}H_{12}N_2O_2$); Anal. Calc. for $C_{16}H_{12}N_2O_2$: C, 72.72; H, 4.58; N, 10.60. Found: C, 72.37; H, 4.51; N, 10.35.

**N-Phthalimidophthalimidine (44)**

A solution of N-carbomethoxyphthalimide (**51**), (6.49 g, 31.6 mmol) in dioxane (110 mL) was added to a solution of hydrazide **118** (4.60 g, 31.0 mmol) in dioxane (15 mL), heated to reflux for 44 h, and filtered. The solution was cooled (11 °C). The precipitate was washed with hexane to remove impurities. The initial filtrate was concentrated *in vacuo* and the resulting residue was washed with chloroform to give a second crop of product. The product was purified by recrystallization from chloroform to give 5.30 g (61%) of **44**, m.p. 257-258 °C; IR ($CHCl_3$) 1795, 1736, 1716, 1370, 708 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 360 MHz) δ 7.97 (m, 3H, *Ar*), 7.84 (m, 2H, *Ar*), 7.67 (m, 1H, *Ar*), 7.54 (m, 2H, *Ar*), 4.81 (s, 2H, $CH_2$); exact mass 278.0691 (278.0691 calc. for $C_{16}H_{10}N_2O_3$); Anal. Calc. for $C_{16}H_{10}N_2O_3$: C, 69.06; H, 3.62; N, 10.07. Found: C, 68.79; H, 3.66; N, 10.00.

**N-Carbobenzyloxyphthalimide (121)**

The procedure of Nefkens *et al*[113] was used. Compound **121** was prepared by a procedure similar to that employed for **51** using (20.0 mL, 0.140 mol) benzyl chloroformate and potassium phthalimide (20.8 g, 0.112 mol) in 67 mL of DMF. Recrystallization from 98% ethanol gave 17.32 g (55%) of **121**, m.p. 114-115.5 °C (lit.[113] m.p. 111 °C); IR ($CHCl_3$) 1806, 1779, 1716, 1308, 1264, 765, 716 $cm^{-1}$; $^1H$

NMR ($CDCl_3$, 80 MHz) δ 7.90 (m, 4H, *Ar*), 7.42 (m, 5H, *Ph*), 5.45 (s, 2H, $CH_2$Ph); exact mass 281.0678 (281.0688 calc. for $C_{16}H_{11}NO_4$); Anal. Calc. for $C_{16}H_{11}NO_4$: C, 68.32; H, 3.94; N, 4.98. Found: C, 68.24; H, 3.88; N, 4.98.

**N-Carbophenoxyphthalimide (122)**

The procedure of Nefkens *et al*[113] was used. Compound **122** was prepared by a procedure analogous to that used for **51** using phenyl chloroformate (12.8 g, 82.0 mmol) and potassium phthalimide (12.1 g, 65.5 mmol) in 40 mL of DMF. Recrystallization form 98% ethanol gave 8.85 g (56%) of **122**, m.p. 144-145 °C (lit.[185] m.p. 142-143 °C); IR ($CHCl_3$) 1809, 1780, 1725, 1313, 1241, 1198, 750, 714 $cm^{-1}$; $^1H$ NMR ($CDCl_3$, 80 MHz) δ 7.95 (m, 4H, *Ar*), 7.39 (m, 5H, *Ph*); exact mass 267.0529 (267.0531 calc. for $C_{15}H_9NO_4$); Anal. Calc. for $C_{15}H_9NO_4$: C, 67.42; H, 3.39; N, 5.24. Found: C, 67.14; H, 3.39; N, 5.33.

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